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THE

Progressive Fish-Culturist

UNITED STATES DEPARTMENT OF THE INTERIOR
FISH AND WILDLIFE SERVICE
BUREAU OF SPORT FISHERIES AND WILDLIFE

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THE HUMBOLDT STATE COLLEGE FISH HATCHERY: AN EXPERIMENT WITH THE COMPLETE RECIRCULATION OF WATER

John W. DeWitt and Ernest O. Salo
Fisheries Department, Humboldt State College
Arcata, California

TO OVERCOME THE DIFFICULTIES PRESENTED BY THE LACK OF A SUITABLE WATER SUPPLY for the operation of a fish hatchery on the Humboldt State College campus, a system which continuously recirculates the water through the hatchery was installed. The general idea of the system and layout was suggested by the senior author, and the detailed plans of construction were prepared by members of the Division of Architecture of the State of California. The junior author has been mainly responsible for the operation of the plant since its completion in 1957.

The system of ponds, raceways, and conduits is permitted to fill with water by gravity flow from a 50,000-gallon storage tank. The tank is filled by flow from a small spring at the minimum rate of about 10 gallons per minute. Once the system is filled, the entire mass of water is set in circulation by the following action: (1) The water is pumped by a 10-horsepower pump through a series of three 230-g.p.m. sand-and-rock filters to an aerating and cooling tower; (2) the water is returned by gravity flow to the hatchery troughs, ponds, and raceways; (3) the water is then either collected in the secondary sump and directed to the primary sump, or collected directly in the primary sump and drawn from it by the pump to complete the cycle.

Initial construction costs and site-development costs were slightly in excess of $100,000. The entire plant covers only one-half acre of land.

Since the summer of 1957 the hatchery has been in constant operation. Five hundred thousand fish and eggs have been handled, and the results of the operation to date are considered successful.

Physical Description and Mechanical Operation of the Plant

Figure 1 shows a general view of the installation. The detailed plan of circulation is presented in figure 2.

When one electric recirculation pump is in operation, the maximum flow to each area is about as follows:

	g.p.m.
Main concrete raceways, each	75
Circular ponds, each--------	35
Earthen pond----------	75
Hatchery troughs, each------	25

Only one of the two electric pumps is kept in operation at any one time, and the pumps are used alternately to provide for uniform wear. An auxiliary gasoline pump is used during emergencies involving power failures. A battery-operated buzzer is used to signal a power failure.

During periods of peak operation it is necessary to backflush all filters approximately once daily. Backflushed water leaves the hatchery system via a sewer line which also carries away water from runoff and other extraneous sources. The backflushing pump is always primed by a water line from the storage tank.

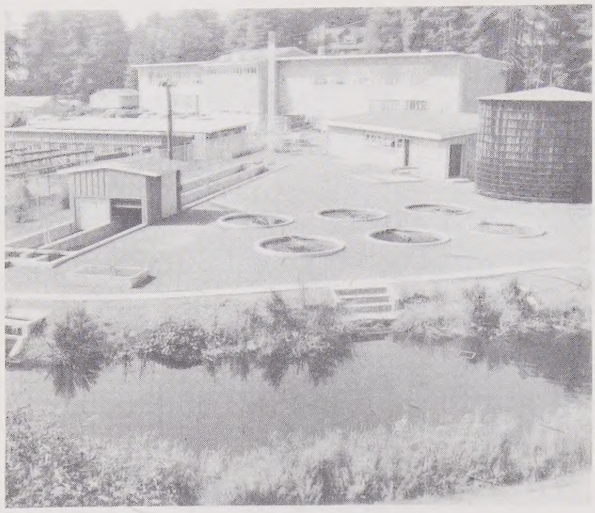

FIGURE 1.-- General view of hatchery and natural resources instructional area. Wildlife building in background.

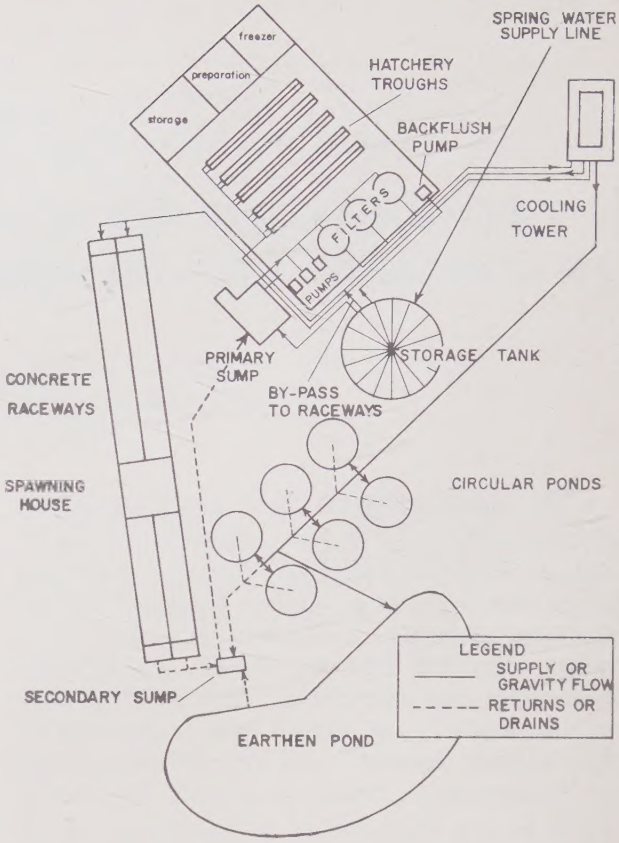

FIGURE 2.-- General hatchery layout and water-circulation pattern.

Water is easily shut off or diverted from place to place in the hatchery by a system of valves. An "alum-pot" is connected to the main flow and--though it has not yet been used extensively--provides a means of adding chemicals to the water at a more or less constant rate for purposes of water clarification, enrichment, or purification.

The storage tank provides the reserve for maintaining the system at full water capacity. When the supply in the storage tank becomes exhausted, a direct line from a city water main provides the water needed for operation. Leakage, evaporation, washing, flushing, and overflows are responsible for losses of water from the system. The spring flow, a minimum of about 10 gallons per minute, has been adequate for continuous operation of the hatchery under all circumstances encountered to date. When any water in excess of that required for operation is present in the system, it merely passes through overflows and down the sewer.

Water Conditions

Water temperatures range from about 46° to 70° F., generally paralleling air temperatures. Dissolved-oxygen concentrations ordinarily range from about 5 to 10 p.p.m. Table 1 shows representative temperatures and dissolved-oxygen conditions at different points in the system during 3 months of the year. During months of warmer weather there is considerable warming of the water as it circulates in the system. The aerating-cooling tower cools the water only 1° or 2° F., on an ordinary summer day.

As the water is passed through any one pond or raceway, there is either a decrease or an increase in the dissolved-oxygen concentration, depending on the number of fish present, algae, food materials, and the time of day. A large concentration of fish in a raceway results in a considerable reduction in dissolved oxygen from the head to the tail end of the raceway, especially during times of feeding. When a large standing crop of algae is in

TABLE 1.--Representative conditions of temperature and dissolved oxygen during March, June, and July, 1959

Month	Location	Temperatures: Water (° F.)	Temperatures: Air (° F.)	Dissolved oxygen (p.p.m.)	Remarks
March	Outlet of earthen pond--	56	------	6.2	
Do.	End of raceway----------	50	------	6.5	Before feeding.
Do.	---do.----------	50	------	5.4	During feeding.
June	Incoming spring water---	52	65	10.0	
Do.	Outlet of earthen pond--	60	65	8.2	Heavy algal growth.
Do.	Input to tower----------	58	65	6.3	After filtration.
Do.	Output from tower-------	58	65	8.9	After aeration.
July[1/]	Incoming spring water---	55	70	9.9	
Do.	Inlet to earthen pond---	63	70	9.3	12:00 a.m., 750 pounds fish.
Do.	Outlet from earthen pond	65	70	7.9	Do.
Do.	Inlet to earthen pond---	64	70	9.0	3:00 a.m.
Do.	Outlet to earthen pond--	65	------	5.8	Do.
Do.	Input to tower----------	66	70	5.4	
Do.	Output from tower-------	62	70	9.4	
Do.	Inlet to raceway--------	65	70	9.5	90 pounds fish present.
Do.	End of raceway----------	66	70	8.6	Before feeding.
Do.	---do.----------	67	70	7.4	After feeding.

[1/] At the time of the determinations, the system contained about 113,000 fish weighing about 1,000 pounds.

the earthen pond, a greater than normal reduction of dissolved oxygen occurs at night and the reduction of oxygen is minimized during the day, even though the pond contains about 300 pounds of fish.

On the basis of the limited analyses made, there appears to be no significant accumulation of dissolved substances in the water, in spite of a retention time of about 4 or 5 days. Table 2 shows representative chemical conditions at times when the hatchery was holding considerable numbers of fish. Frequent backflushing and cleaning of ponds and raceways reduced the extent of accumulation of some substances.

In the winter, even during periods of heavy precipitation, the water in the system usually remained clear enough for good visibility. Actually, the water in the system was sometimes murkiest during the warmer and drier months, when the algal growth was at a maximum and when the spring water supply was at a minimum.

Parasites and Diseases

Fish from several stations have been placed in the hatchery; and even though no special precautions have been taken to prevent the introduction of parasites and disease organisms, the incidence of trouble from these sources has been very low. However, several outbreaks of Ichthyophthirius have occurred. In each outbreak, no great difficulty was experienced in bringing the protozoan under control. Control was obtained by cutting off the flow into and from the pond or trough containing the infested fish, adding the desired amount of chemical, and allowing the treatment to continue for the required time. At the end of the treatment period the normal water flow was restored. The treated water was then allowed to circulate and remain in the system until subsequent dilution, or it was flushed immediately down a sewer line. In some treatments, as with malachite green, the filters trapped most of the material added and it was then backflushed from the system.

TABLE 2.--Representative chemical conditions in the water in March and July 1959

Month	Location	pH	Total solids (mg./l.)	Total alkalinity[1] (mg./l.)	Free CO_2[2] (p.p.m.)
March	Outlet of earthen pond	7.1	83	26	4.5
July	Incoming spring water	6.8	96	32	9.0
Do.	Outlet of earthen pond	6.8	180	38	10.5

[1] All bicarbonate as $CaCO_3$. [2] Determinations by nomograph.

The system lends itself well as an over-sized constant-flow type of chemical-water treatment apparatus, at least for some chemicals. A low concentration of a chemical can be maintained in the entire system over a long period of time while adequate flow and oxygen are also being maintained by the usual means.

Production of Eggs and Fish

Emphasis has been put on the rearing of trout and salmon, but experimental lots of smelt (Hypomesus olidus) and ayu (Plecoglossus altivelis) have been hatched and reared. Approximately 250,000 salmon and steelhead eggs have been shipped into the hatchery, and more than 100,000 eggs have been produced and taken from the brood stock of rainbow and cutthroat trout being maintained in the hatchery. A hatch of about 90 to 95 percent of the eggs was obtained. The survival of the salmon and steelhead to the fry stage was about 95 percent. In addition, 200,000 fish ranging from 960 to 15 per pound have been transferred to the station and reared for varying lengths of time.

The greatest total poundage of fish on hand at any one time was 1,750 pounds in November 1958, but the carrying capacity for that time of year was not reached. During the warmer months the carrying capacity is approximately 1,000 pounds.

Growth rates of the fish have been good and in some cases can be considered excellent. Some typical rates are as follows: Coho salmon averaged 27 per pound 10 months after hatching; without special feeding, rainbow trout grew from 7 per pound in October to 1.2 per pound in March. By the following September, the same fish weighed 1.7 pounds apiece. At 2.5 years of age, the average weight was about 3 pounds.

Acknowledgments

Dr. George H. Allen, assistant professor of fisheries, and Mr. Kenneth A. Johnson, hatchery foreman, provided advice and technical assistance.

A light, easily moved pump is used to increase the oxygen content of ponds at the Ma'agan Michael hatchery in Israel. The pump and motor are attached to a small tin raft that can be floated over even the shallowest areas of the pond. The pumping capacity is 40 cubic meters per hour. The total weight of the apparatus is 100 kilograms. The pump can easily be transferred from pond to pond by one man, and a small tin slide and a vehicle are the only equipment necessary for the task. Water is drawn directly from the pond and can be pumped out again in the form of a single strong stream that extends about 4 meters from the pump or (after the addition of a connecting pipe that divides the volume) in the form of four thin streams that will reach a distance of about 8 meters from the pump.

A COMPARISON OF EGG PRODUCTION OF HATCHERY-REARED BROOK, BROWN, AND RAINBOW TROUT

Keen Buss and Raymond McCreary
Pennsylvania Fish Commission
Bellefonte, Pennsylvania

EACH YEAR AT THE BENNER SPRING FISH RESEARCH STATION, brook trout (Salvelinus fontinalis), brown trout (Salmo trutta), and rainbow trout (Salmo gairdneri) are tagged to establish individuality for use in the selective breeding program. These trout are then held as strains or year classes, this depending on the purpose of study of each lot of trout. Whenever possible, these brood trout are held through more than one spawning season and the accumulated spawning data are analyzed. Normally, comparisons are made among individuals of the same species. It was soon noted, however, that differences among species were very striking, particularly in egg production per pound of brood fish. A knowledge of these differences should be of economic value to both private and public trout hatcheries.

This paper presents information on the volume and number of eggs produced per pound of brood fish. Sufficient detail is included so that comparisons can be made with other locations and trout strains. The analysis is based on the assumption that all other factors are equal, e.g., egg viability, trout strains, and food conversion. It is recognized that these factors, particularly egg viability and trout strains, are not always equal and that they vary from hatchery to hatchery and from State to State. Therefore, the purpose of this analysis is to point out the differences existing in the egg production of the three species of trout at the Benner Spring Fish Research Station so that, with the consideration of other local factors, an intelligent estimate of costs can be made.

Procedure

During the spawning season all sexually mature trout are weighed, measured, and tagged. The female is blotted dry, and her eggs are taken in a dry basin and fertilized. Immediately, before they swell or water-harden, the eggs are placed in a 500-cc. graduate. A rod that has a screen disk attached to its end and is of the same diameter as the inside of the graduate is allowed to settle into the graduate. The body fluids pass through the screen, and a dry measurement of the eggs is obtained and recorded. Then a small sample, usually 50 to 75 cc., is taken and held separately. After eyeing, the small sample is counted manually; and by direct ratio of the volume of the sample to the original lot, the number of eggs is ascertained.

The calculation of egg volume, rather than egg number, per pound of fish provides a better production index because a female may produce a large number of eggs, but they may be abnormally small. This view is supported by correlation studies on brook, brown, and rainbow trout (Wright and Buss, in press) which showed that there is a consistently higher positive correlation between year classes for egg volume than for egg number. Also, egg volume has a higher positive correlation with fish size than egg number.

Group 1

	Two years of age			Three years of age		
	Brook	Rainbow	Brown	Brook	Rainbow	Brown
Average cubic centimeters eggs per pound of fish--	67.9	30.6	76.7	81.5	35.3	96.5
Average number eggs per pound of fish----------	1,509.5	934.4	1,669.1	1,763.0	728.7	1,536.3
Number of individual females tested----------	26	38	8	26	38	24
Average weight of females (pounds)--	1.2	2.8	1.3	2.6	6.2	3.1
Average length of females (inches)--	13.5	18.2	14.5	17.3	24.0	18.9
Average egg size (inches)--	0.164	0.143	0.160	0.173	0.176	0.191
Range: cubic centimeters eggs per pound of fish--	37 - 116	21 - 46	64 - 110	48 - 122	11 - 53	65 - 120
Range: number eggs per pound of fish----------	782 - 2,522	578 - 1,536	1,453 - 2,195	935 - 2,381	200 - 1,174	1,140 - 1,811
Range: weight of females (pounds)--	0.9 - 1.6	2.0 - 4.1	0.9 - 1.8	1.8 - 3.7	4.1 - 10.0	2.1 - 4.1
Range: length of females (inches)--	12.1 - 14.9	16.9 - 20.0	13.4 - 15.9	15.8 - 19.2	20.5 - 27.0	17.3 - 20.1
Range: egg size (inches)--	0.156 - 0.173	0.128 - 0.157	0.156 - 0.165	0.155 - 0.193	0.155 - 0.195	0.185 - 0.203

Group 2

	Three years of age		Four years of age	
	Rainbow	Brown	Rainbow	Brown
Average cubic centimeters eggs per pound of fish-------	35.6	81.2	41.2	81.6
Average number eggs per pound of fish----------	626.1	1,182.1	603.9	870.9
Number of individual females tested----------	44	36	44	36
Average weight of females----------(pounds)--	4.3	3.7	7.7	7.1
Average length of females----------(inches)--	21.1	19.4	25.1	24.0
Average egg size----------(inches)--	0.175	0.195	0.195	0.213
Range: cubic centimeters eggs per pound of fish--------	15 - 70	58 - 112	24 - 61	53 - 127
Range: number eggs per pound of fish----------	350 - 1,020	779 - 1,700	305 - 875	506 - 1,260
Range: weight of females----------(pounds)--	3.3 - 5.8	2.4 - 5.2	5.5 - 10.7	3.6 - 11.4
Range: length of females----------(inches)--	19.6 - 23.1	17.0 - 22.8	22.6 - 27.2	20.9 - 29.2
Range: egg size----------(inches)--	0.155 - 0.190	0.187 - 0.203	0.167 - 0.214	0.195 - 0.230

The egg size is taken during the eyed stage. Eggs are measured in inches by using a 6-inch trough (Lagler 1952). The number of eggs per quart can then be calculated by referring to Von Bayer's table (reproduced, Lagler 1952: 81).

The various analyses concerned with egg production in the three species are presented in the table. These data are for different groups of fish. The rainbow and brown trout in group 1 were selected production fish hatched in the fall of 1952. These same species in group 2 were selected from the production fish hatched in the fall of 1954. The brook trout data are averages of selectively bred strains hatched in the fall of 1954. Normally, at the Pennsylvania State fish hatcheries only brook trout are spawned at 2 years of age. Rainbow trout are not spawned at 2 years because they produce very small eggs with low viability and because they all do not reach sexual maturity at this age. Brown trout at 2 years of age have a low egg viability, and only a small percentage of the females mature at 2 years. The brook trout in the 2-years-of-age section of the table (group 1) are the same individuals as those in the 3-years-of-age section. The same applies to rainbow trout and brown trout in both groups, with the exception that for the brown trout only 8 individuals are recorded at 2 years of age and 24 at 3 years. The discrepancy arises from the fact that only 8 of the 24 were sexually mature at 2 years of age. No records were available for the lots of brook, rainbow, and brown trout included in group 1 at 4 years of age; nor were records available for the rainbow and brown trout at 2 years of age in group 2.

Results and Discussion

Even though rainbow and brown trout are not normally spawned at 2 years of age, it is informative to follow the trend of egg production.

The 2-year-old brown trout produced 2.5 times the volume and 1.8 times the number of eggs that 2-year-old rainbow trout produced. The 2-year-old brook trout out-produced the 2-year-old rainbow trout 2.2 times by volume and 1.6 times by number.

By averaging the egg production of 3-year-old brown trout and rainbow trout in groups 1 and 2, the following results were obtained: 3-year-old brown trout produced 2.5 times as many eggs by volume and 2.0 times as many eggs by number as rainbow trout; 3-year-old brook trout produced 2.3 times as many cubic centimeters of eggs per pound of fish as 3-year-old rainbow trout and 2.6 times as many by number.

At 4 years of age, brown trout (table, group 2) produced almost twice the volume of eggs and 1.4 times the number as rainbow trout. Brown trout eggs are much larger, and this accounts for the lesser number as compared to volume. This also illustrates the advantage of using egg volume as a measure of mass productivity.

As brook trout are normally spawned at 2 years of age and brown and rainbow trout are not, it seems logical to combine the 2-year production of brook trout eggs to estimate the total production from a 3-year-old brook trout. This would be based on the weight of brook trout at 3 years. Brook trout then in their third year of life produced a combined total of 2,456 eggs in number and 113 cc. of eggs per pound of fish. Therefore, because of the 1-year additional spawning, it can be said that, other factors being equal, brook trout eggs are produced at less cost than brown trout eggs, which in turn are produced at half the cost of rainbow trout eggs. This emphasizes the desirability of developing 2-year-old spawning brown and rainbow trout strains which have good egg viability and large egg size.

In conclusion, it can be stated that the economy of holding brood trout can be based not only on age at maturity, egg viability, egg size, and volume of eggs produced per pound of trout, but also on the species. At the Benner Spring Fish Research Station it was found that, at 3 years of age, brook trout are the most productive in volume and number of eggs per pound of trout. Brown trout rank second, and rainbow trout are third with a considerably lower production than the other two species.

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A USEFUL CONTAINER FOR LABORATORY WORK WITH SALMONID EGGS AND FRY

A container developed at the Eastern Fish Disease Laboratory for experimental work on salmonid eggs and fry is inexpensive, simple in design, easily made, and adaptable to several uses.

The unit (figure 1) is made of water-clear methacrylate plastic tubing with a 2-inch diameter and a 0.125-inch wall. The base is a 1.5-inch section with a 2-inch disk of 16-mesh plastic screen cemented across the top with acetate-type glue. The main body is a 4-inch section of tubing cemented to the top of the screened base section. A slot (0.0625-inch) cut through about a third of the circumference, 0.5 inch from the top, permits discharge of water without loss of eggs or fry. Two small holes are drilled near the top so that the unit can be suspended. Cost of materials is about 40 cents a unit.

This unit has been, or can be, used:

1. To simulate a small shipping container or drip incubator, the unit being placed to disperse water coming from a hatchery supply or melting ice.

2. To study toxicity of malachite green to eggs (figure 2).

3. To maintain eggs and fry in separate lots for genetic studies.

4. To study effects of synthetic metabolites on developing eggs.

FIGURE 1.--Egg and fry container made of methacrylate tubing.

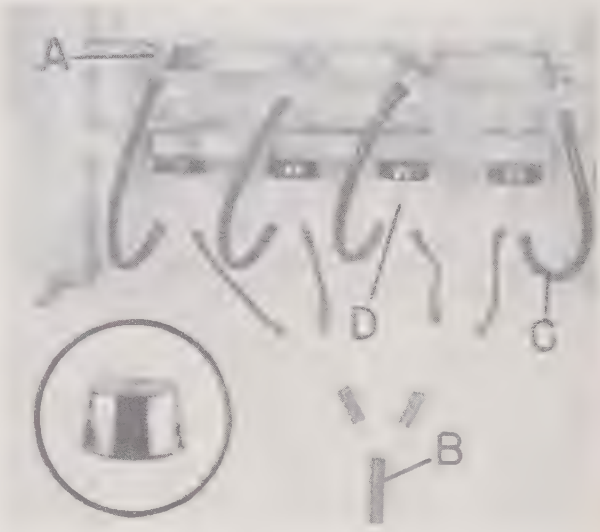

FIGURE 2.--Battery used to determine toxicity of malachite green to eggs. Individual units (A) get water through a common line (B). For exposure to various concentrations, individual supply lines are clamped and units emptied by lowering the gravity drain line (C). After draining is complete, the line is raised and used to introduce the chemical. The inverted Y fitted into the rubber stopper (D) permits ease in operation without disturbing eggs. A 1-inch plastic disk (inset) is fitted a short distance above the inlet to moderate the force of incoming water.

5. To make quantitative and qualitative chemical toxicity studies in flowing or static water.

6. To make studies on the nature and treatment of diseases of eggs and fry.

---KEN WOLF and ROBERT G. PIPER, Bureau of Sport Fisheries and Wildlife, Kearneysville, West Virginia.

HEAVY MORTALITY OF GOLDEN SHINERS DURING HARVEST DUE TO A BACTERIUM OF THE GENUS *AEROMONAS*

William M. Lewis and Michael Bender
Fish Pathology Laboratory and Department of Zoology, Southern Illinois University
Carbondale, Illinois

AEROMONAS is the most important genus of bacterial pathogens of warm-water fishes. Snieszko (1958) recently reviewed the genus with special reference to A. liquefaciens, which he believed to be the organism responsible for most of the fish diseases caused by representatives of the genus. He listed eight scientific names that he considered synonymous with A. liquefaciens. In Bergey's Manual (Breed, Murray, and Smith, 1957), four species of the genus are listed with the following notations: A. liquefaciens, not proved pathogenic to fish; A. punctata, pathogenic to fish; A. hydrophila, septicemic in frogs; and A. salmonicida, pathogenic for fish but nonmotile. Snieszko's list of synonyms (op. cit.) included two of these, A. punctata and A. hydrophila.

Late in the spring, a population of golden shiners (Notemigonus crysoleucas) in a 1/3-acre hatchery pond was partly harvested by seining. Upon being moved to holding tanks, the small and medium-sized fish immediately suffered heavy mortality. Their symptoms included loss of color and swimming at the surface. The next day the larger fish developed saddle-shaped areas of infection around the base of the dorsal fin. Within a week practically all of these fish had died.

A month before the discovery of the disease, 4,000 or 5,000 shiners had been moved from this pond to an adjacent pond. After discovering the occurrence of the disease in pond 1, we suspected that the disease might also be present in the adjacent pond 2.

Two weeks later, pond 2 was seined and the fish were moved to holding tanks. These fish showed the same symptoms as the fish taken from pond 1 earlier and began to die. In addition, the fish from pond 2 showed small hemorrhages scattered over the body, just under the skin. It is possible that the fish from pond 1 also showed this symptom, as the hemorrhages were small and might have been overlooked.

A month later, pond 1 was sampled again; the fish still showed symptoms of the disease. During the period in which the fish were known to harbor the disease, no mortality was observed in the ponds and the fish fed vigorously.

The causative organism was readily isolated from surface lesions and on a number of occasions was isolated from the body cavity and kidney of infected fish. The pure culture was readily transmitted to golden shiners and green sunfish (Lepomis cyanellus) by injections or by inoculated surface abrasions (table 1).

The pathogen grew well on trypticase soy and brain-heart infusion agars. It grew best at 28° C.; no growth occurred above 37° C.

This study was financed by Southern Illinois University and Illinois Department of Conservation.

TABLE 1.--Experimental transmission of a bacterial fish pathogen of the genus Aeromonas obtained from diseased golden shiners

Mode of transmission	Species of fish	Number of fish	Quantity of inoculum (ml.)	Strength of inoculum (millions of organisms per ml.)	Symptoms	Percentage of mortality in 10 days
Injection-----	Golden shiner	5	0.1	5.5	Saddle-shaped discoloration on back and slight lesions on peduncle.	100
Do----------	--do---------	5	.1	.5	Saddle-shaped discoloration on back and small hemorrhages under scales.	40
Do----------	--do---------	10	.1	3.2	---do----------	30
Do----------	--do---------	10	.1	4.9	---do----------	60
Carrier fish[1]	--do---------	5	None	----------	None----------	None
Injection-----	--do---------	10	.1	(2)	Whitish body color and small discolored patches of tissue.	60
Surface abrasion.	--do---------	5	(3)	(4)	Saddle-shaped discoloration and whitish body color.	40
Do----------	--do---------	10	(3)	(4)	Whitish body discoloration----------	20
Do----------	--do---------	10	(5)	(4)	None----------	None
Do----------	--do---------	10	(5)	(4)	---do----------	None
Injection-----	Green sunfish	5	.1	2.0	Centralized surface lesion, but no tissue breakdown; flesh very hard.	100
Do----------	--do---------	5	.1	2.0	---do----------	100
Do----------	--do---------	5	.1	(2)	Body discoloration----------	20
Do----------	--do---------	5	.1	(2)	---do----------	20

[1] To test transmission from fish to fish, 2 shiners were injected with 0.1 ml. of inoculum (50,000 organisms per ml.) and placed in association with 5 normal shiners. The injected fish developed typical symptoms of the disease.
[2] Filtrate. [3] Spray. [4] Not calculated. [5] 10 ml. in water.

The following characteristics place the pathogen in the genus Aeromonas (Breed, Murray, and Smith, 1957): (1) Single polar flagella (Loeffler's flagella stain No. 2) are present; (2) the organism ferments glucose with acid and gas, but slow or no fermentation of lactose occurs; and (3) a negative reaction to the methyl red test is produced. It is not possible to place the organism in any of the four species listed by Breed, Murray, and Smith (1957). Although it most closely resembles A. liquefaciens, there is a difference in that the present form does not produce hydrogen sulfide in either triple-sugar iron agar or Kligler's iron agar.

Detailed morphological characteristics of the pathogen include a size ranging from 0.8 to 1.0 micron in width and 1.7 to 2.0 microns in length. The cells were measured from 48-hour broth cultures and lesions of infected fish. The cells were gram-negative, motile, nonspore-forming, and encapsulated (Manevel's method).

A number of cultural tests were performed on the bacterium to determine its taxonomic position. Liquefaction occurred within 24 hours in a gelatin stab and was crateriform to saccate. Complete liquefaction occurred within 5 days. Nitrates were reduced to nitrites, and indole was produced. Growth on blood agar was heavy, but no hemolysis was observed. The medium was discolored (greenish brown) after 48 hours. The Voges Priskauer test was negative after 2 days of growth. Starch was hydrolyzed (no color with iodine); only slight growth occurred on potato agar. Urea was not attacked. Milk was partly digested with a change to acid.

A test of the pathogen's sensitivity to various bactericides demonstrated that it was particularly sensitive to aureomycin, Chloromycetin, Terramycin, and tetracyclin (table 2).

TABLE 2.--Bactericidal sensitivity of a bacterium, Aeromonas, pathogenic to golden shiners

[Plates were inoculated by adding 0.15 ml. of washed bacterial culture. The growth was heavy at 24 hours (time of measurement).]

Bactericide	Concentration (mcg.)	Zone of inhibition[1] (mm.)
Aureomycin	30	10
Bacitracin	10	0
Carliomysin	15	0
Chloromycetin	30	14
Erythromycin	15	5
Neomycin	30	3
Penicillin	[2]10	0
Polymyxin B	300	4
Terramycin	30	12
Tetracyclin	30	12
Triple Sulfa	1,000	0
Dihydrostreptomycin	10	1

[1] Zone of inhibition was measured from the edge of the disc to the edge of zone.

[2] Penicillin is measured in units.

Discussion

The purpose of this report is to supplement known occurrences and add to our knowledge of the etiology of pathogens of the important group Aeromonas. The limits of observations relating to warm-water fish pathogens are such as to make it difficult to gain much information about the importance, nature, and control of this group of pathogens. The senior author has repeatedly observed heavy spring mortality in warm-water fish populations. These mortalities have exhibited symptoms of bacterial infections; but without the kind of study reported here, the exact cause of this type of mortality cannot be accurately established.

As already noted, it was not possible to establish positively the specific name of the pathogen. It fits the description of the genus Aeromonas very well, but it does not completely fit the description of any of the four species listed in Bergey's Manual (Breed, Murray, and Smith, 1957). However, the organism fits Snieszko's list of characteristics for the identification of A. liquefaciens (Snieszko 1958). It is possible that the present pathogen is a variant of A. liquefaciens.

Snieszko (1958) considered A. liquefaciens to be the causative agent of "red leg" in frogs. In the hatchery from which the infected shiners described in this report were obtained, "red leg" is common at times in the leopard frog (Rana pipiens).

Outbreaks of aeromonal infections appear to occur most commonly in the spring. Snieszko (1958) attributed this seasonal occurrence to the fish's reduced disease resistance, the result of winter dormancy and starvation.

The chronic nature of the infection in the shiner was of particular interest to the authors. It appeared that as long as the shiners were in the pond, they did not die of the infection; nevertheless, in a period of 2 months they did not recover. When fish were harvested, they immediately showed symptoms of the disease and mortality commenced.

Acknowledgments

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TOXICITY OF PYRIDYLMERCURIC ACETATE TO CUTTHROAT TROUT

In the summer of 1959 pyridylmercuric acetate was tested on cutthroat trout at the newly opened National Fish Hatchery at Jackson, Wyoming. At this station the water is of moderate hardness, has a pH of 7.6, and at the time of this experimental work had a temperature of 49° F.

Four 2-year-old cutthroat trout were put into a metal tank and treated for 1 hour with PMA at a concentration of 1:500,000. Showing no distress, these fish were returned to the rearing tanks, which contained 1,300 fish of the same age and size (1.4 per pound). The next morning all of these fish were treated in the tank for the same period and at the same concentration. Though the fish showed no distress during the treatment, 2 hours later they were in distress and losses began. In 24 hours the lot had suffered 24 percent mortality.

Several days later, 3- to 18-inch lake, brown, brook, rainbow, and cutthroat trout were placed in a tank and given the same treatment with PMA. The larger cutthroat were fish that had survived the previous treatment. At the end of 3 days, the lake, brown, brook, and large cutthroat trout showed 100 percent survival, and the rainbow trout showed 100 percent mortality. The cutthroat trout fingerlings that had not been treated previously had suffered almost 25 percent mortality. Most of the losses occurred within the first 24 hours.

The cutthroat trout treated were of the Snake River strain, and taxonomic studies by Dr. A. D. Welander of the University of Washington have indicated that these fish have not hybridized with the rainbow. Nevertheless, it was observed that these cutthroat were subject to mortality after the initial treatment with PMA but immune to the second, and that the greater part of the deaths occurred among the larger, healthier, more highly colored fish.

--- GLEN L. HAMMER, Bureau of Sport Fisheries and Wildlife, Cortland, New York.

FIVE-YEAR REPORT ON THE DEVELOPMENT OF THE FISHERY OF A 25,000-ACRE DOMESTIC WATER SUPPLY RESERVOIR IN MASSACHUSETTS

Robert S. McCaig, James W. Mullan, and Cheney O. Dodge
Massachusetts Division of Fisheries and Game
Westboro, Massachusetts

QUABBIN RESERVOIR is the largest and most important water unit in Massachusetts. It constitutes the main water supply for Metropolitan Boston and represents approximately 33 percent of the water area available to the angling public. Created by the completion of a dam on the Swift River in 1939, the reservoir reached spillway level in 1946, when it had maximum and average depths of 150 and 50 feet, respectively.

Fishing was prohibited until 1946, when 46 miles of the reservoir's 118 miles of shoreline were opened to shore fishing only. (See figure.) In the spring of 1952 boat fishing was initiated. Three State boat liveries were constructed; facilities included boat and motor rentals, launching sites for private boats, and parking. A fee was charged for use of these facilities. Records of these transactions included the number of shore fishing parties, the number of parties using private boats, and the number of parties renting State boats, as well as the number of anglers in the last two categories. A record of departure and return was maintained on both State-owned and private boats.

A creel census, begun in 1954, has been maintained.

This work was undertaken with Federal Aid in Fish Restoration funds, being part of Dingell-Johnson Project F-6-R, Massachusetts.

Methods of Study

No boats could be used except those launched, or rented, at one of the three liveries. Fishing pressure on "walk-in" gates, barricaded against ingress by cars, was assessed by car counts. Creel sampling was executed by stratified schedules. Sampling effort involved 5 man-days a week (except for 3 man-days a week in 1956) between the third Saturday in April and middle or late October, during which the Quabbin was opened to boat fishing. Shore fishing is permitted beyond the closing date for boats, but--as ice fishing is prohibited--is limited by ice cover and inclement weather.

Field data were expanded through use of the Metropolitan District Commission (M. D. C.) records kept in relation to boat usage from an annual sample, ranging from 19 to 24 percent, of the recorded boat fishermen (table 1).

Shore-fishermen data were expanded similarly, with the exception that car counts made at "walk-in" gates, and the estimated number of fishermen per car, were added to the total supplied by the M. D. C. for shore fishermen at "drive-in" areas. Number of anglers interviewed ranged from 5.5 to 0.4 percent (table 1). Data from the day shore fishermen are less reliable than the data from the boat fishermen, owing to the smaller sample interviewed. Comparisons between the two categories of data, however, show

fair agreement. Furthermore, should any bias be inherent, it lies in expansion of the pan-fish and not the game-fish catch.

A third group, night shore fishermen, was handled as a separate entity. These fishermen primarily fished for and caught brown bullheads. All the cars counted at "walk-in" gates after dark were grouped in this category. Boat fishing and use of parking facilities at the "drive-in" areas after dark are prohibited, a condition differentiating this group. These data are the least reliable, representing very limited sampling (from 0.7 to 2.4 percent of the estimated numbers of night fishermen). Any distortion of the total harvest through use of these data affects principally the brown bullhead.

Fishing Pressure, Harvest, and Stocking

It was estimated that 216,408 fishing trips comprising 1,158,436 angling hours, in which 591,471 fish weighing 211,734 pounds were harvested, were made at Quabbin Reservoir in the period 1954-58 (table 2). On the basis of total reservoir area, annual fishing pressure and harvest amounted to 9.3 fisherman hours and 4.7 fish weighing 1.7 pounds per acre. On the basis of the area open to fishing (16,850 acres; see figure), 13.7 hours were spent in harvesting 7 fish weighing 2.5 pounds per acre.

Fishing effort declined from 45,448 angler trips in 1954 to 32,297 in 1955, after which it rose to slightly less than the 1954 level by 1957. In 1958, fishing effort increased to 60,360 angler trips (table 2).

The maximum catch of 156,753 fish (53,069 pounds) was recorded in 1954. Numerically the harvest in 1958 ranked lowest with 98,931 fish taken, whereas by weight it ranked second with 47,573 pounds harvested (table 2). This is reflected in an annual decrease in the average catch of 0.70 fish per hour in 1954 to 0.29 in 1958,

TABLE 1.--Fishing pressure and harvest by categories of anglers--boat fishermen, day shore fishermen, and night shore fishermen--for the period 1954-58 on Quabbin Reservoir

	1954	1955	1956	1957	1958
Boat fishermen					
Total angler trips	21,848	19,454	21,818	23,279	36,658
Total hours	142,154	125,109	137,768	149,684	246,527
Total fish harvested	88,113	71,990	71,389	53,188	63,723
Total pounds	29,052	23,918	22,702	21,788	33,113
Average angler day (hours)	6.5	6.4	6.3	6.4	6.7
Fish per hour	0.62	0.58	0.52	0.35	0.26
Pounds per hour	0.20	0.19	0.16	0.15	0.13
Number fishermen checked	5,256	4,524	4,236	5,045	6,846
Percent checked	23.7	23.3	19.4	21.7	18.7
Day shore fishermen					
Total angler trips	13,949	8,364	12,401	13,822	18,118
Total hours	52,699	31,003	57,124	62,475	68,965
Total fish harvested	34,100	18,800	42,709	21,625	21,073
Total pounds	13,739	5,886	13,443	7,696	8,323
Average angler day (hours)	3.8	3.7	4.6	4.5	3.8
Fish per hour	0.65	0.61	0.75	0.34	0.31
Pounds per hour	0.26	0.19	0.24	0.12	0.12
Number fishermen checked	632	520	47	114	496
Percent checked	4.5	5.5	0.4	0.8	2.7
Night shore fishermen					
Total angler trips	9,651	[1] 4,479	4,894	6,568	5,584
Total hours	28,921	-------	17,697	17,931	20,379
Total fish harvested	34,540	-------	15,206	40,857	14,135
Total pounds	10,279	-------	3,662	12,000	6,137
Average angler day (hours)	3.0	-------	3.6	2.7	3.7
Fish per hour	1.19	-------	0.86	2.2	0.69
Pounds per hour	0.36	-------	0.21	0.80	0.30
Number fishermen checked	232	-------	56	48	128
Percent checked	2.4	-------	1.1	0.7	2.3

[1] 1955 success figures not available.

and in an increase in the average weight of fish in 1958 (0.48 pound) over the norms of the previous years (0.31 to 0.36 pound).

In 1954, game fish--trout, bass, and pickerel--formed 2.1 percent of the numbers and 11.1 percent of the weight of the harvest. By 1958, these values had risen to 5.4 and 23.0 percent, respectively. Pan fish [1] --principally the yellow perch, chain pickerel (Esox niger), brown trout (Salmo trutta), rainbow trout (Salmo gairdneri), brook trout (Salvelinus fontinalis), lake trout (Salvelinus namaycush), brown bullhead (Ameiurus nebulosus), black crappie (Pomoxis nigro-maculatus), white perch (Morone americana), yellow perch (Perca flavescens), bluegill (Lepomis macrochirus), pumpkinseed (Lepomis auritus), rock bass (Ambloplites rupestris), white sucker (Catostomus commersonni), fallfish (Leucosomus corporalis), smelt (Osmerus mordax), and walleye (Stizostedion vitreum).

[1] Largemouth bass (Micropterus salmoides), smallmouth bass (Micropterus dolomieu),

TABLE 2.--Summary of total fishing pressure and harvest on Quabbin Reservoir during the period 1954-58

	1954	1/ 1955	1956	1957	1958	Total or average
Total angler trips	45,448	27,818	39,113	43,669	60,360	216,408
Total hours	223,774	156,112	212,589	230,090	335,871	1,158,436
Average angler day (hours)	4.9	5.6	5.4	5.3	5.6	5.4
Total fish harvested	156,753	90,792	129,325	115,670	98,931	591,471
Total pounds	53,069	29,794	39,814	41,484	47,573	211,734
Fish per hour	0.70	0.58	0.61	0.50	0.29	0.51
Pounds per hour	0.24	0.19	0.19	0.18	0.14	0.18

1/ Does not include 4,479 estimated night shore fishermen, for whom there were no success figures.

white perch, and brown bullhead--constituted 70 to 89 percent of the harvest by number and weight annually.

Less stability of population is indicated for two minor pan fish. The harvest of black crappie totaled 3,189 in 1954, but only 850 were taken in 1955, and even fewer were taken in subsequent years (table 3). Average size increased from 0.34 pound in 1954 to 0.61 pound in 1958, indicating a real decline in population density (table 4). On the other hand, the harvest of the rock bass seemed to be increasing. None appeared in the catch prior to 1957 (when 157 were taken), but 85 were taken in 1958.

The catch of white suckers dropped from 2,863 in 1954 to 821 in 1955, and no white suckers were taken in subsequent years (table 3). Whether the catch records are reflective of population abundance, again, is problematical. It has been demonstrated that Massachusetts anglers are primarily interested in game species and least interested in species such as white suckers (Stroud 1952, Stroud and Bitzer 1955). Spawning suckers concentrated at two of the three launching sites for boats. Accessibility and novelty possibly accounted for some of the catch of this species in the initial years these areas were open.

Spring bass fishing in Massachusetts was initiated on the Quabbin Reservoir in 1954. Prior to that year, the bass season coincided with the July 1 State-wide opening. An average of 65 percent of the season's take of bass occurred during the normally closed period.

Under the extended season, the catch of largemouth bass increased yearly, except for a slight decrease in 1955 (table 3). The take of smallmouth bass declined for 2 years after 1954, when the maximum harvest of 689 fish was recorded. Thereafter, the catch of smallmouth bass rose, regaining by 1958 about half of its previous high. The main reservoir possesses ideal habitat for smallmouth bass: the bottom type is rock and rubble, etc. Nevertheless, smallmouth bass populations are almost exclusively confined to the 547-acre Pottapaug Pond area, which is isolated by a baffle dam at boat mooring area No. 3. (See figure.) Abundance of largemouth bass in this area appears comparable to that in the main reservoir. There appeared to be an increase in the catch of largemouth bass from the Pottapaug area, an increase paralleling that of the main reservoir. If so, substitution of largemouth bass for smallmouth, to some degree, might be accountable for the change noted. Unfavorable population balance in Pottapaug, complicated by the presence of a large white perch population that is slower-growing than that existing in the main reservoir, undoubtedly is also a factor.

Although the chain pickerel is only of intermediate importance in the overall fisheries, its relative abundance, as

TABLE 3.--Total angler harvest, by species, from Quabbin Reservoir during the period 1954-58

Species	1954		1955		1956		1957		1958	
	Number	Pounds	Number	Pounds	Number	Pounds	Number	Pounds	Number	Pounds
Largemouth bass----	1,576	3,079	1,390	2,414	1,804	2,051	1,860	2,527	2,062	3,144
Smallmouth bass----	689	998	169	309	118	169	249	319	299	524
Chain pickerel-----	997	1,605	573	780	3,279	3,476	743	981	853	1,414
Brown trout--------	98	183	112	169	41	65	1,054	1,183	395	1,514
Rainbow trout------	77	47	20	14	5	5	647	837	368	808
Brook trout--------	4	4	23	8	36	5	902	503	11	12
Lake trout---------	-------	------	------	------	57	57	636	1,565	1,340	4,964
Brown bullhead-----	63,162	18,840	28,236	9,322	52,026	15,461	60,659	19,654	36,985	16,274
Black crappie------	3,189	1,092	850	451	846	453	392	175	667	407
White perch--------	38,984	13,417	29,297	9,104	34,116	11,055	25,601	9,033	25,892	12,167
Yellow perch-------	31,756	6,093	21,911	4,254	25,361	4,371	16,117	3,334	24,691	4,939
Sunfish[1/]----------	13,877	2,694	7,271	1,335	11,610	2,639	6,616	1,303	5,250	1,352
Rock bass----------	-------	------	------	------	-------	------	157	45	85	21
White sucker-------	2,863	4,979	821	1,583	-------	------	-------	------	------	------
Fallfish-----------	53	38	12	12	5	2	37	25	33	33
Total----------	156,753	53,069	90,792	29,794	129,304	39,808	115,670	41,484	98,931	47,573

[1/] Includes the bluegill, pumpkinseed, and yellowbelly sunfish.

reflected in both the catch and population sampling, is surprising. Muddy shallows, supporting aquatic vegetation, generally conceded to be essential for pickerel reproduction, are significantly lacking in any visual appraisal of the reservoir. Instead, the shoreline is precipitous, rocky, and wind-lashed. Undoubtedly, the catch records mask the true abundance of the pickerel. Open-water fishing for pickerel has been demonstrated (Stroud and Bitzer 1955) to be relatively ineffective in accounting for a substantial portion of the legal population. Of greater significance, the harvest records depict the normal yearly fluctuations in catch of this species and the importance of dominant year classes. For example, in 1956 there was a sixfold increase in the catch of pickerel over the previous year, and this increase was followed in 1957 by an almost equally drastic decline (table 3).

TABLE 4.--Average weight (in pounds) of fish caught at Quabbin Reservoir

Species	1954	1955	1956	1957	1958
Largemouth bass--	1.95	1.74	1.13	1.36	1.52
Smallmouth bass--	1.45	1.83	1.43	1.28	1.75
Chain pickerel---	1.61	1.36	1.06	1.32	1.65
Brown trout-----	1.86	1.51	1.59	1.12	3.83
Rainbow trout----	.61	.70	1.0	1.29	2.20
Brook trout------	1.0	.35	.14	.56	1.09
Lake trout-------	----	----	1.0	2.46	3.70
Brown bullhead---	.30	.33	.30	.32	.44
Black crappie----	.34	.53	.54	.45	.61
White perch------	.34	.31	.32	.35	.47
Yellow perch-----	.19	.19	.17	.21	.20
Sunfish----------	.19	.18	.22	.20	.26
Average-------	.34	.33	.31	.36	.48

Between 1952 and 1957, the reservoir was stocked with 290,137 lake trout, mostly fingerlings. The lake trout first appeared in 1956, when 67 (average, 1 pound) were caught. Ten of these were returned as being under the 18-inch size limit. The following year (1957), 636 lake trout (1,565 pounds) were harvested. These fish averaged a little under 2.5 pounds. Also reported were 51 lake trout of less than legal size. In 1958, 1,340 lake trout (4,964 pounds) were harvested (table 3) and 621 of less than legal size were reported. Average weight of legal lake trout was 3.7 pounds, the range being from 2 to 6.5 pounds. Fish up to 8.5 pounds were reliably reported, although not recorded in the census.

Whether this species has spawned successfully in the reservoir is not known. Though a few mature fish could have been present in the fall of 1957, a large number of mature lake trout were known to be present in the fall of 1958.

Although brook, brown, and rainbow trout were landed yearly before stocking in 1957, the limited population apparently did not offer sufficient inducement for large numbers of anglers to employ methods (principally deep trolling) suitable to taking such fish. Small natural populations of brook and brown trout are to be expected because tributary stream habitat is limited. Rainbow trout do not reproduce in the reservoir, and the few caught before 1957 originated from hatchery fish stocked in a few of the headwater tributaries. The problem is further complicated by the fact that the native brook trout dominates in the tributaries, yet does not significantly contribute to the reservoir fisheries. Most of the brook trout reported were taken in the confluences of the tributaries with the reservoir.

The first stocking of 1,000 each of fin-clipped 9- to 12-inch brook, brown, and rainbow trout was made on an experimental basis in mid-March 1957 (table 5). The first-year returns were 66, 23, and 36 percent, respectively. Second-year returns of the brown and rainbow trout were 10 and 14 percent, respectively; there were no returns of the brook trout.

In 1958, in the closed section of the reservoir, 1,000 brown and 1,000 rainbow trout were stocked. These trout realized first-year returns of 2.4 and 10.1 percent, respectively (table 5).

The peak harvest of brook, brown, and rainbow trout occurred in 1957, when 2,603 were taken. This was correlated with the first catchable stockings of these species and the first significant influx of lake trout into the catch. However, it is not to be overlooked that about half of this catch consisted of unmarked trout. In 1958, though the overall catch was just under a third of the previous year, the proportionate relationship between the marked hatchery and unmarked trout remained about the same.

Aside from the lake trout, introductions of two other exotics have been made. Just over 10,000,000 fry, 3,045 subadult, and 990 adult walleyes were introduced in the period 1953-58. The fate of these introductions is a mystery. Three walleyes originating from fry stockings were sampled in an overnight gill net set in the summer of 1955. One of the adults stocked from Lake Erie in 1954 was taken by an angler in the summer of 1958. Reports of anglers catching about six under-sized walleyes and the sighting of one adult at night in 1958 provide the only other leads to the survival of walleye introductions.

Smelt were first introduced in this reservoir in the spring of 1953, when 2,000 small (2- to 4-inch) fresh-water individuals were stocked. In 1954, 40,000 similar fish and 3,000 sea-run adults were stocked in the reservoir. Descendants of these stocks have established a huge population, first observed in characteristic spawning runs in the spring of 1957. They have continued to increase, spawning in tributary streams and in shoal areas. By 1958, they were so abundant as to inhibit water distribution from the reservoir by clogging intake screens and flow meters.

TABLE 5.--Summary of 9- to 12-inch fin-clipped brook, brown, and rainbow trout stocked and harvested at Quabbin Reservoir, 1957 and 1958

Trout	Date stocked	Total number (and pounds)	1957 harvest: Number (and percent)	1957 harvest: Pounds	1958 harvest: Number (and percent)	1958 harvest: Pounds	Total: Number (and percent)	Total: Pounds
	1957							
Brook----	March 15	1,000 (489)	662 (66.2)	351	--- ------	------	662 (66.2)	351
Brown----	--do----	1,000 (436)	226 (22.6)	210	102 (10.2)	353	328 (32.8)	563
Rainbow--	--do----	1,000 (429)	361 (36.1)	303	144 (14.4)	433	505 (50.5)	736
	1958							
Brown----	March 15	1,000 (461)	--- ------	---	24 (2.4)	37	24 (2.4)	37
Rainbow--	--do----	1,000 (345)	--- ------	---	101 (10.1)	81	101 (10.1)	81

Discussion

The success of the trout and smelt plantings is responsible for the increase in fishing pressure and decline in overall catch rate in recent years. Initial fishing pressure was triggered to a large extent by novelty and dropped off when realizations failed to meet expectations. This trend was reversed when the lake trout began to enter the catch. Correspondingly, the numerical catch rate suffered as more of the total effort was expended in pursuit of fewer but larger fish, particularly trout.

An abundance of smelt played an integral role in the equation, being responsible for the early attainment of legal size of the lake trout. Before the establishment of a significant smelt population, the lake trout reached 8.6, 12.5, and 15.6 inches at 2, 3, and 4 years of age, respectively. With a plentiful supply of

smelt, lengths of 10.6, 14.5, and 18.3 inches are now attained for the same respective years of life. Rapid and more accelerated growth of each succeeding year class of stocked lake trout parallels the continued buildup in the smelt population, even in the light of increased population density of all trout.

The same relationship is borne out by an analysis of the growth of 146 wild brown trout. Before the establishment of a significant smelt population in 1956, brown trout averaged 13.8, 15.7, and 19.7 inches at 3, 4, and 5 years of age, respectively. Lengths of 16.6, 18.4, and 21.5 inches are now attained for the same respective ages.

Likewise, there was no radical difference in the growth of the 9- to 12-inch brown and rainbow trout stocked in 1957 between identical plantings made in 1958. Brown and rainbow trout averaged about a fourfold gain in weight after 6 months in the reservoir both years. Brown and rainbow trout carryovers from the 1957 stockings averaged 3.8 and 3.0 pounds, respectively, when taken in 1958. Some individual brown trout weighed 6 to 7 pounds 16 months after liberation. It can only be concluded that observable intraspecific and interspecific competition have not yet set in for any of the trout in the Quabbin.

Accelerated growth may be a partial explanation for the increased catch of unmarked brown and rainbow trout. More than likely, recruitment has not been significantly altered, but mortality perhaps has, with the end result that more of these fish are reaching trophy size before succumbing to natural causes.

A marked increase in the average size of warm-water species with piscivorous dietary habits harvested in 1958 could possibly be interpreted on a cause-and-effect relationship attributable to smelt. Lack of spatial overlap, at least during most of the growing season, regulates against such a blanket assumption. The white perch harvested during 1958, while maintaining their relative numerical contribution to the total catch with other years, averaged 0.47 pound, as compared with their averages between 0.31 and 0.35 pound for the 4 previous years (table 4). Strangely enough, the bullheads showed a similar phenomenon. Yellow perch, many of which do overlap in their depth distribution with smelt, averaged about one-fifth of a pound during all 5 years of the census. Growth data on white perch partially suggest that some of the larger white perch may be responding to this abundant forage supply. Spatial overlap of this pelagic species with smelt is known, and many of the white perch examined in 1958 had eaten large numbers of smelt. However, the whole problem of the effects of smelt on the white perch and the other warm-water species is complicated and possibly can be unraveled only with the passage of time.

Adverse effects of the smelt in interfering with water distribution from this and connected reservoirs are serious. Gravity-fed systems from principal distribution heads are impaired by smelt-clogged screens and subsequent loss of pressure. The M.D.C. hopes to relieve this condition by control of smelt spawning runs in tributaries. What influence this will exert on the future fisheries is problematical. However, the problem does attest, along with the success achieved from the trout planting, to the value of stocking in the absence of severe competition.

Two-thirds of the volume of Quabbin Reservoir consists of cold-water habitat. The warm-water species of the reservoir were not adapted to exploiting this environment to maximum advantage, whereas the introduced cold-water species--trout and smelt--were. Conversely, it appears that the walleye introductions encountered exactly opposite conditions of prior occupancy, by a complicated warm-water species complex, of the available habitat niches to which they are suited.

In conclusion, future prospects for the fisheries are bright, with the exception of the conflict of interest over smelt. It still remains, however, that the primary use or value of the Quabbin is as a water supply. Increased predatory pressure on the smelt through expansion of game-fish

stocks will be emphasized, as a result, in an attempt to turn a liability into an asset. The validity of the latter has been demonstrated by the small-scale stockings of brown and rainbow trout, which have realized returns exceeding 100 percent on a weight basis. Conceivably, excessive stockings of brown and rainbow trout could depress the growth of lake trout through competition for the same food (in this case, smelt). Such an occurrence naturally should be avoided. The creation of brown and rainbow trout fishing is definitely subordinate to lake trout fishing. This is predicated on the fact that the lake trout have the potential of establishing a significant, self-supporting big game fishery that does not involve expensive maintenance.

Acknowledgment

Sincere appreciation is expressed to Mr. J. A. Boccardy (now of the Bureau of Sport Fisheries and Wildlife), who was the Quabbin Reservoir project leader in the years 1954-55.

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------ and Harold Bitzer

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A SPECIAL MOUNT FOR AN OUTBOARD MOTOR

With a 5.5-horsepower outboard motor mounted 6 inches above the normal position and the tilt-adjusting bar of the motor set on release, a 12-foot aluminum boat can be used in very shallow water.

The mount is made of 1.75- by 9- by 15-inch wood (fir), painted to match the boat. The mount is attached to the stern with four 0.25-inch stove bolts, fastened down with wing nuts. Flat washers are used on both ends of the bolts. For easier insertion and removal of the bolts, the holes in the stern are slightly larger than the diameter of the bolts. Short 0.25-inch bolts and nuts are used to plug holes below the water line after the mount is taken off.

Because of turbulent air around the propeller, only low speeds can be maintained with the motor raised. For higher speeds in deeper water, the motor must be in the normal position. To change without going ashore, the motor is taken off the mount,

the mount is detached, bolts are inserted as plugs (if necessary) below the water line, and the motor is attached to the stern in the normal position. A safety chain is attached to the motor at all times.

---RUDY STINAUER, State Fish Hatchery, Pecatonica, Illinois.

IMPROVEMENTS IN A MICROPROJECTOR FOR FISH SCALES

Elmer S. Phillips, Department of Extension Teaching and Information
and
Dwight A. Webster, Department of Conservation
Cornell University, Ithaca, New York

MANY FISHERY INVESTIGATIONS require that considerable time be expended in determining the ages of fish. Usually this involves the examination of the projected images of scales. Often the specimens run into the thousands, so that any improvement that will increase efficiency or lessen fatigue of the scale reader is important. One of the earliest, and apparently still one of the best, basic designs for a scale projector was described by Van Oosten, Deason, and Jobes (1934). Projectors based on this design are now commercially available.

Prior to the construction of a new scale projector for use in fishery biology at Cornell University, possible modifications in optical arrangements of this basic design were considered. Several changes were made in the apparatus described (op. cit.) or in its commercially available counterpart. These included (1) greater comfort by increasing leg room under the projection screen; (2) redesign of the optical and mirror system to permit greater ease in placing scale specimens and in focusing; (3) use of a fixed objective and movable specimen stage for focusing, permitting accurate reproduction of magnifications for any given lens system; and (4) addition of an interchangeable back-projection surface, permitting the substitution of a standard photographic plate-holder, and thus allowing use of the apparatus for making photographic negatives or prints directly. Comments on the last two possibilities were made by Lagler (1956).

The projector described here has been in use for 4 years and has given excellent results.

Optical and Projection System

The optical system was an adaptation of the Bausch and Lomb VH Micro-Projector. This was equipped with the 32- and 48-mm., f4.5 iris-diaphragm Micro-Tessar lenses and appropriate condensers, and a 16-mm. fixed-aperture objective with appropriate condenser. The transformer for alternating current was also used. The microscope tube, supporting legs, and circular light screen usually supplied with this unit were not purchased.

The arrangement of the optical parts is such that the lenses are mounted in a standard position, hence giving consistent magnifications on the viewing screen. Focusing is accomplished by moving the specimen into the focal plane of the lens. In this projector, magnification for each lens measured at the center of the image is: 16 mm., 84.65x; 32 mm., 40.50x; 48 mm., 26.75x.

Recently the multiple nosepiece that fits the microscope mount was purchased to hold all three lenses. This further increases the efficiency of the unit by allowing any lens to be moved quickly into position. It is advisable to purchase the nosepiece at the time the unit is constructed. A later purchase necessitates building a special short-coupling to maintain the same magnifications derived from the single-lens mount.

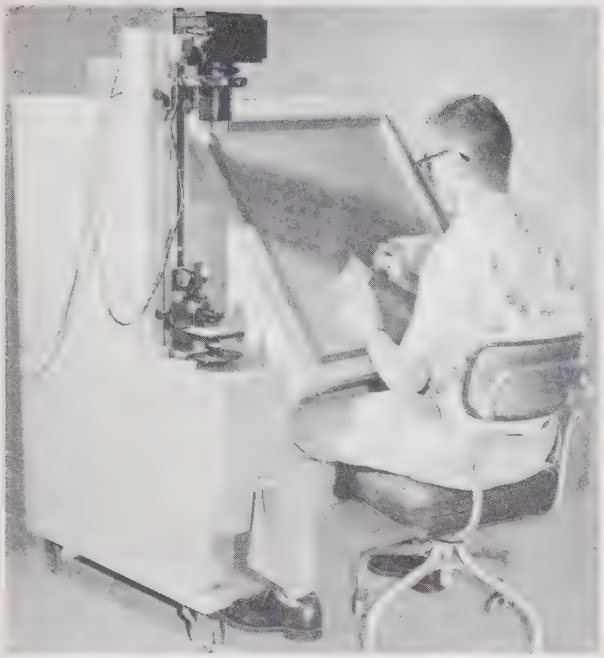

FIGURE 1.--Finished scale projector.

The arrangement of mirrors results in a shift of approximately 45° in the axial orientation of the image. A guide that is easily attached to the projector stage automatically orients the scales consistently (provided, of course, that the scales are oriented consistently in mounting). When plastic impressions are used, the imprinted side should be placed down on the stage to give the sharpest image.

The image under observation must be viewed at a right angle from the surface to eliminate distortion due to parallax. This is the normal viewing position; hence it works no hardship when scales are to be measured.

Construction of the Unit

Because the scale plans are necessarily involved, it will be easier to follow the construction of the unit by first studying figure 1 (the completed unit before the addition of the multiple nosepiece) and figure 2 (the scale model furnished the carpenter's shop along with the plans). The scale plans are shown in figure 3.

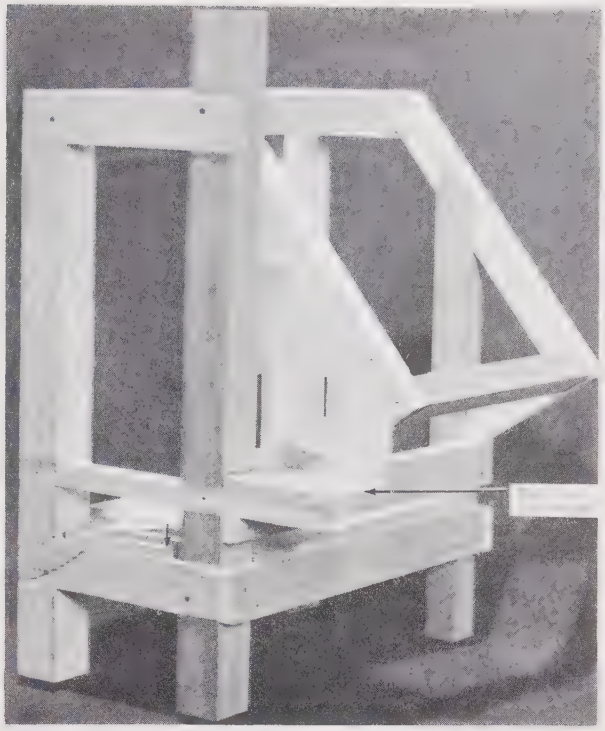

FIGURE 2.--Scale model of a microprojector submitted to the carpenter's shop with the plans. Slight modifications in the joint construction were made by the cabinetmaker to assure strength.

The basic construction of the unit is wood; three 4 by 4's and one 3 by 3 piece are used as vertical supporting elements. The 3 by 3 post is to the right of the operator and clearly seen in figure 2. The floor of the unit is 12.5 inches from the room floor and made of 0.75-inch plywood to which is glued 0.125-inch tempered masonite for additional strength. A similar piece of plywood is used to mount the first mirror 1 foot 6 inches from the floor.

The frame of the unit is horizontally supported by 2 by 4's glued and screwed in place. The angle of the receiving screen is 50°. The entire skeleton of the structure is sheathed in 0.25-inch plywood; with the exception of the back (which is screwed in place to facilitate changes or adjustments), these pieces are also glued and nailed. The viewing screen is mounted in a frame hinged at the top (figure 1).

SCALE PROJECTOR

Well seasoned wood. Joints bolted and glued. Cover all sides with 1/4 " plywood screwed or nailed except back which is held with screws only. Hinge front frame at top.

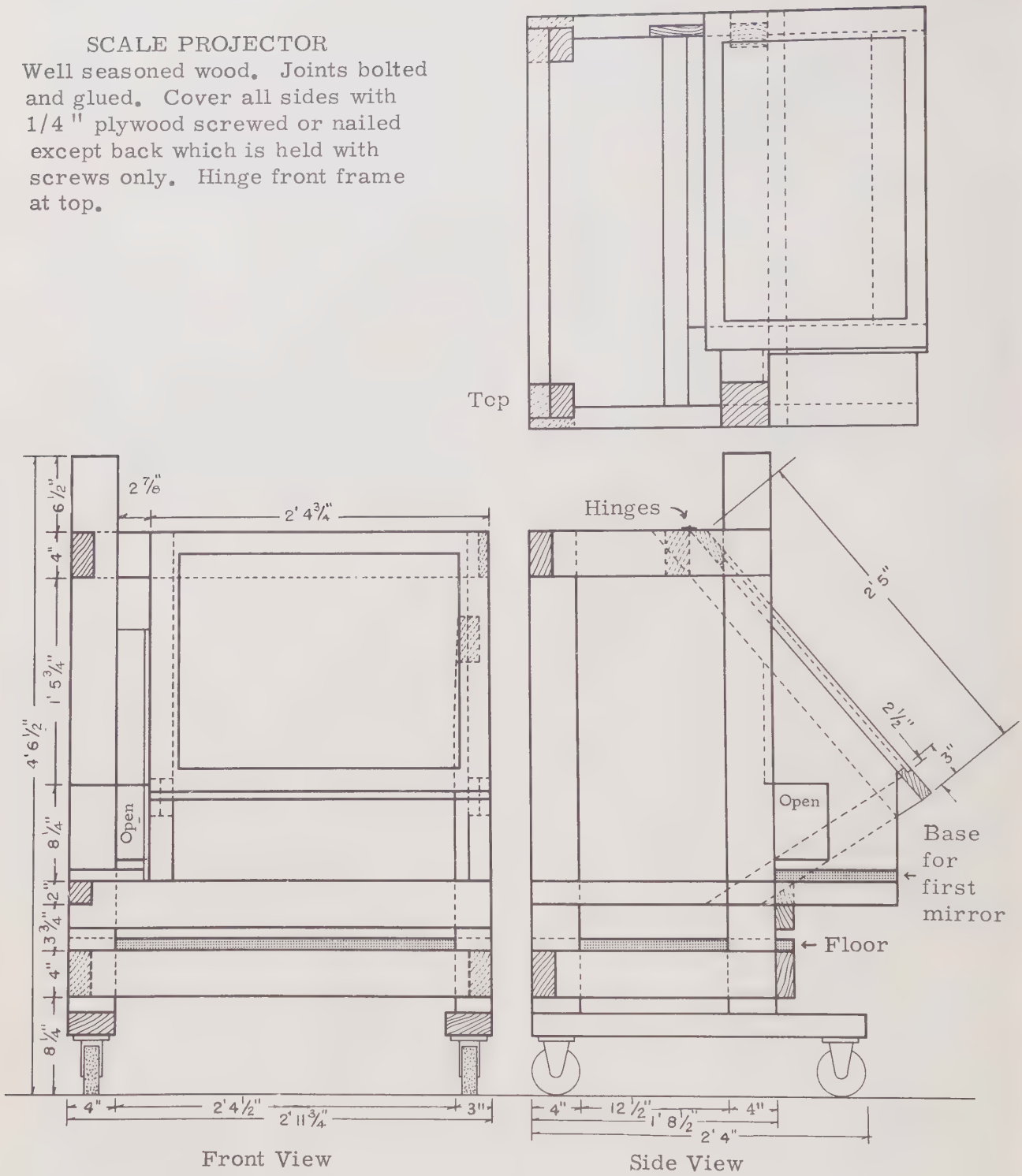

FIGURE 3.--Scale plans of microprojector as submitted to the carpenter's shop.

A three-sided hollow box of 0.5-inch plywood (seen just below the lens in figure 1) that covers the first mirror slides into grooves, screws firmly in place, and allows complete removal of the first mirror for cleaning or replacement if necessary. A sponge-rubber ring insulates the box from the base of the optical equipment to prevent dust from filtering onto the first mirror. A drawer below the first mirror housing is used to store lenses, condensers, test slides, lens papers, and similar material.

With this construction, knee room approaches 12 inches in depth from the edge of the screen. Placing the bottom of the unit as indicated leaves adequate foot room.

An added refinement evident in the photograph is the large rubber casters that can be locked in position. A 2 by 4, 2 feet 4 inches long, was fitted to the bottom of the four main supports to retain the dimensional heights indicated. This entailed shortening each support to accommodate the casters plus the 2 by 4's.

Mounting the Optical Equipment

The wedge-shaped metal track furnished with the microprojector is mounted on the longest 4 by 4 as follows:

1. The center of the microprojector track is mounted vertically 1 inch from the righthand side of the 4 by 4 with two 3-inch flathead screws. New holes may have to be drilled and countersunk to accommodate the screws.

2. Between the track and the 4 by 4, place two small pieces of 0.125-inch wood, about 2 inches long, to elevate the track from the 4 by 4.

3. The bottom of the microprojector track should be adjusted so that when the bracket that holds the lens in position strikes the pin built into the base by the manufacturer, 0.0625 inch of clearance is maintained between the cubical box and the bracket holding the lens. It is at this point that the sponge-rubber ring will be inserted. The pin in the track of the microprojector should be approximately 7.25 inches from the base that holds the first mirror. This places the axis of the specimen slide about 2 feet 6 inches above the floor of the room for the 32-mm. lens.

The first silver-surfaced mirror, 3.5 by 5 inches on 0.5-inch glass, is mounted under the lens in a yoke so that horizontal and vertical adjustments (tilt and side swing) can be made easily. The image is then reflected to a larger mirror, 16 inches square on 0.25-inch glass, similarly mounted to reflect the image to the transilluminated screen (figure 4).

Wood or metal yokes may be made to hold the mirrors (A and B, figures 4 and 5). However, two requirements must be included in the construction. Each yoke must be fastened with bolts, or screws, to allow vertical pivoting at the surface of the mirror. The single screw or bolt also shown in the base of the yoke (X, figure 5) must be perfectly centered below the horizontal center of the vertical pivoting motion (front view, figure 5).

The pivot-point height for the first mirror (A', figure 4) is 4.5 inches above the heavy masonite-covered base; this means that the mirror pivots off its vertical center (2 inches of the mirror extends above the pivot point, and 3 inches extends below the pivot point). This is necessary to accommodate the image size disparity which results when all parts of the image are not equidistant from the lens.

4. Although the first mirror (A', figure 4) can be centered by measurements derived from the drawings, it may be more accurate to place the mirror in relation to the beam of light from the microprojector. To do this, align the filament, condenser, and lens. A quick and accurate method is to cover the lens condenser with tissue or lens paper and adjust the filament and condenser in the lamp housing so that the image of the filament is centered on the paper over the condenser.

Remove the paper and project the beam of light to the base used to support the first mirror. Place an X on the masonite in the exact center of the light beam. This marks the horizontal pivot axis for the yoke of the first mirror and centers the screw (or bolt) position for the first mirror (X, figure 5).

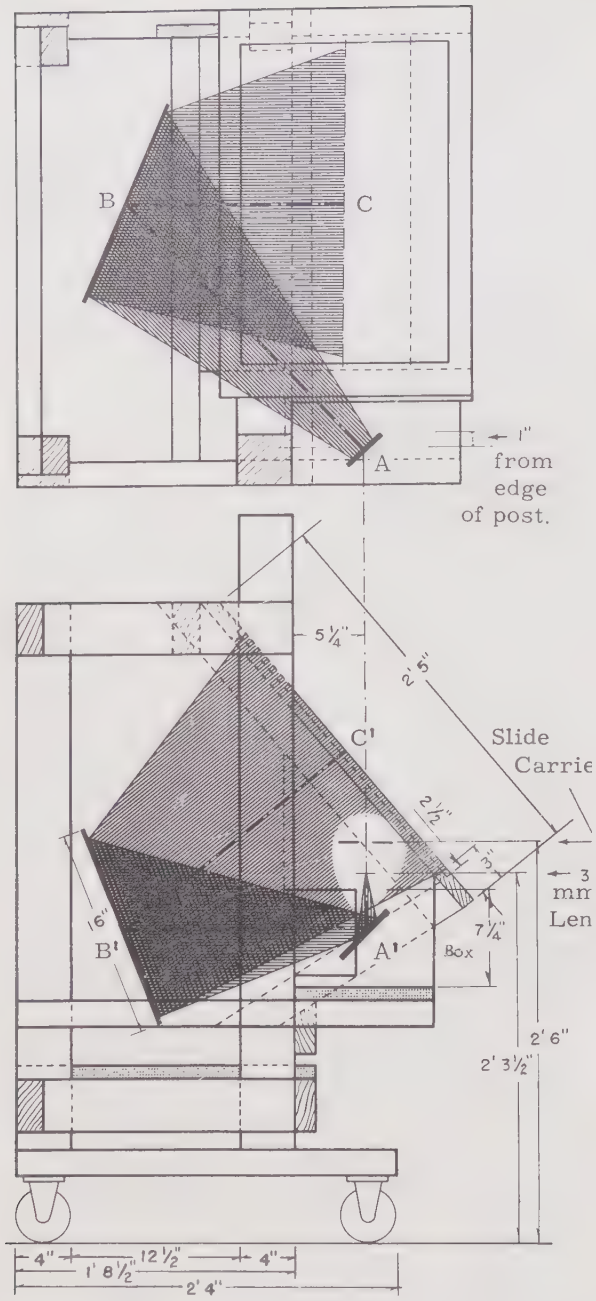

FIGURE 4.-- Schematic diagram of optical plans superimposed over the top and side views of the construction plans.

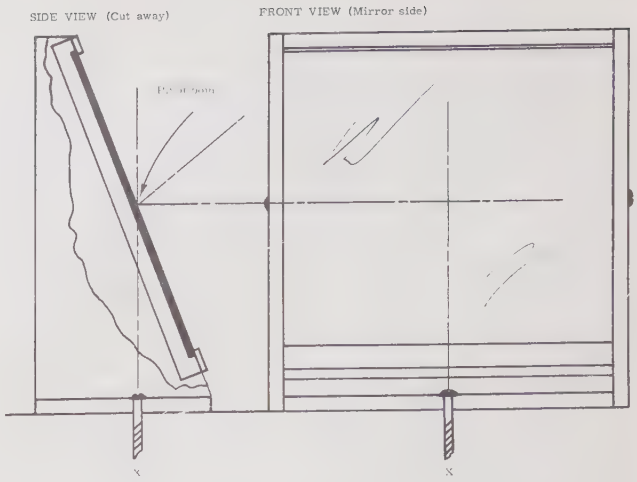

FIGURE 5.--Diagram of yoke construction and mirror alignment.

5. Scribe a + in the center of a piece of plastic such as is used to receive the scale impressions. Securely tape this to the projector stage so that the image of the + centers on the X made on the masonite base of the first mirror. Keep the plastic in place for further adjustment.

6. Drill a hole to accommodate the screw that holds the base and install the first mirror.

7. To place the second (16-inch) mirror, mark on the floor a line drawn 90° from the center of the viewing screen (B-C, figure 4). This should be parallel to the sides of the frame of the projector.

The vertical pivot point for the second mirror is 10 inches from the projector floor. Magnification can be adjusted by moving the second mirror nearer to or farther from the screen on axis B-C (figure 4). However, if it is moved in either direction, the 10-inch-high pivot point will change on an extension of the center line B'-C' (figure 4).

8. Temporarily place a white cardboard, of the same thickness and size as the mirror, into the yoke that ultimately will hold the second mirror. The center of the cardboard should be marked with a dot.

9. Tilt the first mirror in either or both planes until the projected + is perfectly centered on the dot. Tighten the centering screws of the first mirror so that it will not move.

10. Replace the cardboard with the second (16-inch) mirror.

11. Temporarily holding the yoke of the second mirror in place, repeat operation No. 9 with the second mirror until the + centers on the viewing screen.

12. Use a carpenter's square or similar tool to test the angle of the line B'-C'; it should be 90° from the screen. It is important that the angles of the lines B-C and B'-C' both be 90° from the screen. If this principle is not met, magnifications on the screen will not be uniform.

13. Center, drill, and tighten the centering screw of the second mirror to the floor. Lock each mirror to its respective floor with a second bolt or screw, some distance from the centering screw.

Several materials suitable for the back-projection screen were tested, including ground glass, matt plastic, latex, and ektalite. Ektalite gave a sharp image but was not available in a large-enough size. Kodak Day-View-Screen (White Type) was selected as most satisfactory. The back-projection screen was mounted with the receiving or image-forming side inside the unit to protect it from abrasion.

Adaptation for Photography

Permanent records of scales are frequently useful for purposes of comparison or for illustrations. These can be made with this projector by substituting for the viewing screen a sheet of 0.75-inch plywood fitted with a conventional 8- by 10-inch camera back. This has the usual ground glass for focusing and takes standard 8- by 10-inch film holders. Instead of film, however, the holders may be loaded with photographic paper. Additional prints of high quality can be made using the developed print as a paper negative.

If critical measurements are to be made on the prints, papers that do not change size, such as Resisto Rapid, should be used. There may be some difficulty in standardizing exposure when scales of varying sizes or variations in the depth of plastic impressions are involved. Unfortunately, most standard exposure meters are not sufficiently sensitive in the light intensity involved to be of use.

General Comments

The cost of the projector will be nearly $1,000. Of this, the major portion will be spent for optical parts (about $600). Other relatively fixed costs are mirrors (about $50) and viewing screen ($25). Labor for cabinetwork will vary according to local conditions; it was estimated at $100 to $200 by the Carpenter Shop at Cornell University in 1955. Installation and alignment of the optical parts and mirrors will be an additional expenditure.

The entire unit could be built of metal, which would slightly reduce the size of the instrument. Building of wood calls for a word of caution: use well-seasoned wood and a waterproof seal or paint. The inside of the unit should be painted dull black.

The completed projector is a reasonably precise piece of optical equipment and should be treated accordingly. This comment, which may seem superfluous, was prompted by observing the condition of a similar projector after being transported in a pickup truck.

Acknowledgments

This scale projector was constructed with funds from a fishery research grant by Mr. and Mrs. Donald P. Ross of Montchanin, Del. Mirrors for this model were furnished by courtesy of Evaporated Metal Films Corporation, Ithaca, N. Y. Several students made helpful suggestions for more convenient operation of the projector. Mr. C. W. Lynn, Jr., fishery technician, Cornell University, completed the finishing work. Dr. A. W. Eipper reviewed and commented upon the first draft of the manuscript.

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NEUTRALIZING CHLORINE IN CITY WATER FOR USE IN FISH-DISTRIBUTION TANKS

Earl A. Pyle
Bureau of Sport Fisheries and Wildlife
McNenny National Fish Hatchery, Spearfish, South Dakota

DISEASE-PRODUCING ORGANISMS IN WATER may be effectively controlled by adding chlorine in an amount somewhat greater than that sufficient to oxidize all the organic matter. The chlorine that remains is called residual chlorine and serves as an indication that the water has been made safe as far as bacteria are concerned. Most municipal water supplies are treated with chlorine added as gaseous chlorine or as chloramine (a combination of chlorine and ammonia). Water containing 0.5 p.p.m. residual of free chlorine is generally considered safe for domestic use (Garver 1955). It is well known, however, that the amount of residual chlorine present in water may differ greatly among cities and at times may also vary considerably from the same source. The average daily chlorine content of the Carbondale, Illinois, water supply was approximately 2.1 p.p.m., ranging from 1.2 to 3.1 p.p.m. (Cole and Lewis 1958).

Chlorine is considered one of the best disinfecting agents for hatchery sanitation. However, fish-culturists are well aware that even small amounts of chlorine in water can be lethal to fish, and extreme care must be exercised when using it.

Although published accounts could not be found, there have been verbal reports of mortality occurring among transported fish after the emergency use of municipal water in a distribution tank or even after using ice made from municipal water. Circumstantial evidence in each case indicated that residual chlorine could have been the cause of mortality. Ellis (1937) reported that 1.0 p.p.m. residual chlorine killed goldfish in 96 hours. Rodgers (1949) reported serious losses among displays of live fish when chlorinated city water was used.

Sodium thiosulfate ($Na_2S_2O_3 . 5H_2O$) is a highly soluble chemical of low toxicity which will neutralize chlorine in solution. It is stable, inexpensive, and may be obtained at drug or camera-supply stores. The reaction is represented by the following equation:

$$2Na_2S_2O_3+Cl_2 \rightarrow Na_2S_4O_6+2NaCl.$$

If this chemical could be used at a concentration exceeding that required to neutralize even the most highly chlorinated water and not in itself be toxic to fish, a supply could be carried in the distribution unit as a "first aid" remedy in an emergency.

This report describes three experiments which were conducted to accomplish these objectives:

1. To determine the toxicity, to some of the commonly propagated sport fishes, of water containing graded amounts of residual chlorine comparable to that which might be found in municipal water supplies.

2. To determine the tolerance levels of these fishes to various solutions of sodium thiosulfate.

This paper was prepared while Mr. Pyle was attending the Leetown In-service Training School at Kearneysville, West Virginia, from September 1958 to August 1959.

3. To determine the effect, if any, that the products of neutralization of chlorine with sodium thiosulfate have on these fishes, and the efficiency with which the reaction occurs.

Materials and Equipment

Eastern brook trout (Salvelinus fontinalis), brown trout (Salmo trutta), rainbow trout (Salmo gairdneri) fingerlings, and smallmouth bass (Micropterus dolomieu) fingerlings were randomly selected as test fish. The trout averaged 7 centimeters in length and the bass 4.5 centimeters in length. Cylindrical 4-liter glass jars, each supplied with compressed air, were used as containers for the trout. To maintain uniform temperature, the jars were kept immersed in flowing water having a temperature of 12.5° C. (54° F.). The smallmouth bass were kept in stainless-steel troughs, and the ratio of bass to the volume of water was such that it was unnecessary to introduce supplementary oxygen. Observations were made over a 24-hour period, and the fish were not fed during the experiments.

Effects of Chlorine on Fish

Five brook trout were added to each of four jars containing 2.5 liters of water with 0, 0.5, 1.0, and 1.5 p.p.m. chlorine obtained from commercial Clorox (active ingredient, sodium hypochlorite 5.25 percent by weight). Similarly, five bass were added to each of four stainless-steel tanks containing 100 liters of water with the same concentrations of chlorine. The tests were made in duplicate. The concentration of each solution was verified by the ortho-tolidine method in a Hellige comparator.

Effects of Sodium Thiosulfate on Fish

This experiment differs from the preceding experiment only in that 0-, 1-, 5-, and 10-percent solutions of technical sodium thiosulfate were used. For reasons of economy, the volumes of solution were reduced, i.e., 1 liter each for the trout and 20 liters each for the bass. Rainbow trout and brown trout were tested in the same manner as the brook trout, and the results were so nearly identical that it was not considered necessary to include them.

Neutralizing Chlorine with Sodium Thiosulfate and the Effect on Fish

It was attempted to simulate conditions which might be experienced on an actual distribution trip. Five randomly selected brook trout were added to each of four jars, in duplicate, containing 500 milliliters of water with compressed air introduced in the smallest possible amount. The fish were held in this environment for 2 hours to allow an accumulation of metabolic wastes which conceivably could have a bearing on the results. At the expiration of this period, technical sodium thiosulfate was added to three jars at 1, 5, and 10 times the amount needed to neutralize 1.5 p.p.m. residual chlorine. Immediately 2 liters of water containing 1.5 p.p.m. residual chlorine was added to each of the three jars that contained the sodium thiosulfate solutions. The fourth jar received 2 liters of water which contained no chlorine; this jar served as a control. Determinations made immediately by the ortho-tolidine method demonstrated that no discernible chlorine was present in any of the test jars, thus indicating the rapidity of the reaction.

Over a 24-hour period no mortality occurred and the fish exhibited no signs of distress.

Discussion

Median mortality among brook trout and smallmouth bass occurred within a few hours in concentrations of chlorine within permissible limits of municipal water systems (table 1). It is entirely possible, therefore, that mortalities could occur as a result of transporting fish in water from a municipal supply.

Tolerance of the test fish to technical sodium thiosulfate was very high (table 2). Exposure to a 5-percent solution caused

TABLE 1.--Occurrence, in time, of distress and median mortality for brook trout and smallmouth bass in various concentrations of chlorine over a 24-hour period

Concentrations (p.p.m.)	Brook trout		Smallmouth bass	
	Distress	Median mortality	Distress	Median mortality
0 (Controls) ---	None	None	None	None
.5 ----------	1 hour	90 minutes	6 hours	15 hours
1.0 ----------	20 minutes	50 minutes	4 hours	6 hours
1.5 ----------	10 minutes	40 minutes	3 hours	5 hours

TABLE 2.--Occurrence, in time, of distress and median mortality for brook trout and smallmouth bass in various concentrations of technical sodium thiosulfate over a 24-hour period

Concentrations (p.p.m.)	Brook trout		Smallmouth bass	
	Distress	Median mortality	Distress	Median mortality
0 (Controls) ----	None	None	None	None
1 ----------	None	None	None	None
5 ----------	3 hours	5 to 19 hours[1/]	7 hours	7 to 23 hours[1/]
10 ----------	40 minutes	70 minutes	40 minutes	90 minutes

[1/] These figures are not precise because of an interruption in the observations. For the purposes of this paper, however, the information is considered adequate.

no distress among brook, rainbow, and brown trout fingerlings over a 3-hour period; the smallmouth bass exhibited no distress for 7 hours. All fish survived for 24 hours and apparently were not affected by a 1-percent solution. A 1-percent solution (10,000 p. p. m.) is 5,000 times as strong as is needed to neutralize 1 p. p. m. chlorine.

Results obtained from these experiments indicate that an amount of sodium thiosulfate, adequate to eliminate the chlorine in any municipal water and with no adverse effects on the fish, could easily be carried in the distribution unit, immediately available if needed.

Recommendations

For each 100-gallon volume of the distribution tank, 20 grams of technical sodium thiosulfate should be carried in the distribution unit to neutralize the chlorine in the water from any municipal source. This will furnish a large margin of safety, with no toxic effects on the fish. To avoid exposing the fish to chlorinated water, the sodium thiosulfate should be added prior to the introduction of the water.

A polyethylene container for the sodium thiosulfate is ideal for this purpose.

Summary

1. Solutions of chlorine within permissible limits of municipal water systems are toxic to fingerling brook trout and to fingerling smallmouth bass.

2. Brook, rainbow, and brown trout fingerlings and smallmouth bass fingerlings exhibited no distress over a 24-hour period when exposed to a 1-percent solution of sodium thiosulfate. Exposure to a 5-percent solution caused no distress in the test trout for 3 hours or in the smallmouth bass for 7 hours, though subsequent mortality occurred in all. Median mortality occurred within 90 minutes among all fish exposed to a 10-percent solution.

3. Sodium thiosulfate applied at these rates will rapidly and effectively neutralize water containing 1.5 p.p.m. chlorine.

4. Products of the neutralization of chlorine with technical sodium thiosulfate were nontoxic to the test fish.

5. A fish-distribution unit can carry 20 grams of technical sodium thiosulfate for each 100 gallons of the tank for use in neutralizing chlorine should it become necessary to use water from a municipal source.

Acknowledgments

The author wishes to thank Mr. W. E. Purcell and Mr. F. J. Howard, trainees at the Leetown In-service Training School, September 1958 to August 1959, for their assistance in conducting the experiments.

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A MECHANICAL FISH LOADER

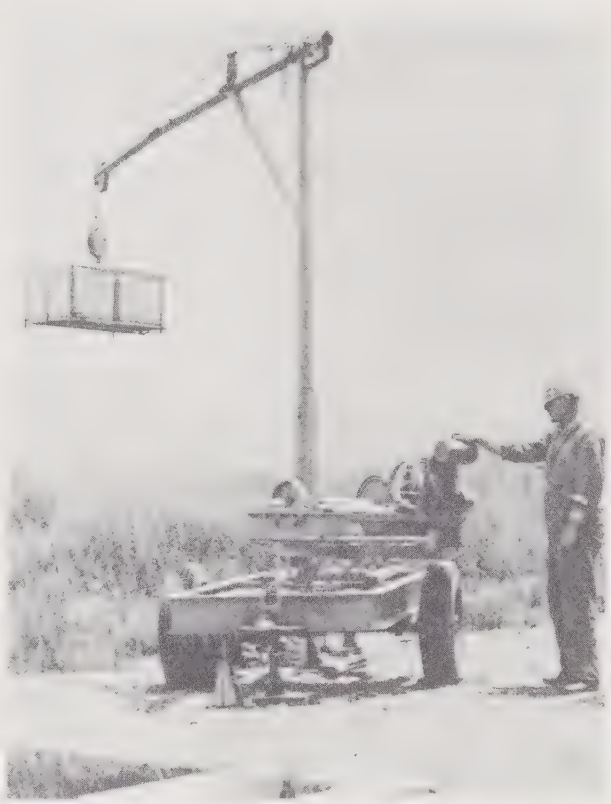
This mechanical fish loader is mounted on a two-wheel trailer that can be towed by a small tractor or pickup truck.

Powered by a 6-horsepower gasoline motor and heavy-duty winch with a 24-inch drum, the loading arm or boom is on an inverted truck hub and can be pivoted to any position. The boom is telescoped so that its reach can be adjusted. The cable that lifts the basket is threaded through the pipe with pulleys at each end.

The basket--steel-reinforced, perforated aluminum--has a trap bottom that is operated by a spring catch for quick opening and closing. It holds 200 pounds of fish.

Scales attached to the basket-lifting cable are set at zero, with the weight of the basket, and protected by a chain yoke and boomer. The weight of the fish can be read quickly by releasing the boomer on the protective yoke.

---TOM E. FRENCH and RAYMOND F. JOHNSON, Bureau of Sport Fisheries and Wildlife, Leadville, Colorado.



PHARMACOLOGICAL STUDIES ON THE MELANOPHORES IN *FUNDULUS HETEROCLITUS*

Charles G. Wilber
Comparative Physiology Branch, U. S. Army Chemical Warfare Laboratories
Army Chemical Center, Maryland

IT HAS BEEN KNOWN FOR SOME TIME that acetylcholine disperses the melanophores of Fundulus and renders the fish dark in color (Parker 1948). In order to get this effect, it is necessary to eserinize the fish to prevent the rapid destruction of the injected acetylcholine. In specimens of Fundulus so prepared, an injection of acetylcholine (in the range of 1 to 100 p.p.m.) results in a darkening of the fish in 20 to 30 minutes.

In general, the response of melanophores in Fundulus to drugs can be summarized as follows: "Drugs which stimulate the central or the sympathetic nervous system cause a contraction of the melanophores through stimulation of their pigment-motor mechanism and those which have a depressant effect cause an expansion." (Wyman 1924: 125.)

One fruitful method for obtaining information on fundamental physiological mechanisms in man is the comparative method. Much basic work on nerve transmission has been accomplished on cold-blooded forms. Fish are a convenient group of cold-blooded animals to work with because of ease of handling and care. Moreover, they respire in a liquid medium. Hence the testing of concentration and exposure time is less tedious with fish than with animals respiring in air.

There is a large background of information on the physiology of melanophores in Fundulus. It therefore seemed worthwhile to study the response of these melanophores to decamethonium, hexamethonium, and octamethyl pyrophosphoramide (OMPA), all drugs of interest to the Chemical Corps.

Materials and Method

The systemic insecticide OMPA that was used in this work was obtained from the Monsanto Chemical Company: Sample No. 52-12-112. The label indicated that the sample contained 70 percent OMPA, 20 percent other phosphates, and 10 percent inert substances. No enzymatic standardization of the material was done.

Decamethonium bromide (C - 10) and hexamethonium bromide (C - 6), also used in this work, were obtained from the Burroughs Welcome Co., Inc., of Tuckahoe, New York.

General characteristics of these compounds are given in the sixth edition of the Merck Index. The physiological and pharmacological effects of methonium compounds on mammals have been discussed by Paton and Zaimis (1952). The effects of OMPA on mammals have been reported by DuBois, Doull, and Coon (1950).

The killifish, Fundulus heteroclitus, was used in this work. Many previous investigators have used this species as a test animal in making studies of melanophore responses in fish (Parker 1932, 1948; Wyman 1924). It seemed desirable to use the same species in the present work to facilitate the comparison of these results with those already in the literature.

The fish were injected intraperitoneally[1] (by means of a 26-gauge needle) with the several drugs or exposed to them in sea-water solutions. Ten fish were used in each experimental and control group. Experimental and control aquaria were set over white backgrounds with a light shining from above. This procedure causes untreated fish to be light colored.

Results

A mixture containing 3 grams per liter of commercial OMPA in sea water was prepared. Light-colored Fundulus were prepared by exposing several dozen of the fish in aquaria over a white background under a bright light for 24 hours. The melanophores were in stage-1 condition (Parker 1948). The light fish were put into the OMPA mixture in large glass aquaria. Controls were exposed similarly in pure sea water. At the end of 40 minutes, all of the experimentally immersed fish were dark and the control fish were still light. After a 2-hour exposure all fish were alive: the experimental fish were very black; the control fish were light. The experimental fish remained dark until death; all were dead after 7 hours of exposure.

A second group of light Fundulus were injected with OMPA. One milliliter of commercial OMPA was mixed with 20 milliliters of sea water. Each fish received 0.5 milliliter of this mixture or 0.025 milliliter of commercial OMPA.[2] Control fish received 0.5 milliliter of sea water. Sixty seconds after the experimental fish had been injected, a dark black band developed around the fish at the level of the injection. At the end of 10 minutes, the band was widened both anteriorly and posteriorly. At the end of 60 minutes, all fish were patchy with black. Shortly thereafter all experimental fish were completely black. The melanophores were in stage 3 (Parker 1948). No melanophore response was observed in the control fish.

A group of experimental fish were immersed in a sea-water solution of hexamethonium bromide, 400 milligrams per liter. Although observations were made regularly for 4 hours, no changes were apparent; the experimental and the control fish were the same color at the end of 4 hours as at the start of the experiment.

A solution of hexamethonium bromide in distilled water was injected into 10 light Fundulus (0.5 milliliter of a solution containing 10 milligrams of C-6 per milliliter). Within 60 seconds, all the fish were black.

A solution of decamethonium bromide of the same concentration was similarly injected into 10 light Fundulus. They all remained light. Consequently, 10 dark Fundulus (prepared by keeping them over a black background) were selected and injected with the same amount of C-10. Within 60 seconds, all of the fish had lightened to a pale yellow.

In order to ascertain whether the present procedure would give results similar to those reported in the literature (Wyman 1924), each of 10 Fundulus was injected with 0.5 milliliter of a 0.5-percent solution of atropine sulfate in distilled water. Although the control fish injected with pure water showed no change, the experimental fish were completely black in about 2.5 minutes. If such darkened fish were given an additional injection of C-10 (5 milligrams in 0.5 milliliter of distilled water) or of distilled water, no further changes in color were induced.

Finally, 10 fish were immersed in a solution of decamethonium in sea water (400 milligrams per liter). After 4 hours of immersion, no changes in these fish were observed. The table on the following page summarizes these results.

[1] By inspection, no visible leakage took place. As precise toxicity values were not the object of this work, the matter was not pursued further. Because of the large dose, an excess of OMPA was in the fish after injection even if some leakage took place.

[2] This amount was equivalent to 1/60 milliliter of pure OMPA because the commercial sample contained only 70 percent OMPA.

RESPONSE OF FUNDULUS TO DRUGS

Agent and pathway	Fish color	Melanophore condition
OMPA: injection	Darkened	Dispersed.
OMPA: immersion	Darkened	Dispersed.
C-10: injection	Lightened	Concentrated.
C-10: immersion	No effect	----------
C-6 : injection	Darkened	Dispersed.
C-6 : immersion	No effect	----------

Discussion

It has been fairly well established that the dominant mechanism of melanophore control in Fundulus is nervous (Prosser 1950). Furthermore, the weight of published evidence leads to the view that the nerve fibers causing dispersal of melanophores in Fundulus exert their action through acetylcholine (Parker 1940, 1948).

In the present work OMPA rendered light killifish dark. It is known that OMPA has very little anticholinesterase activity in vitro and must be metabolized in the liver to produce an anticholinesterase product (DuBois, Doull, and Coon, 1950). It seems evident, therefore, that Fundulus has the capacity to convert OMPA into an anticholinesterase product which brings about the accumulation of acetylcholine in the fish. The excess acetylcholine acts on the melanophores, dispersing their pigment and darkening the fish.

Apparently OMPA is readily absorbed by fish through the gills or the skin, for darkening resulted after immersion as well as after injection. Melanophore-dispersing substances, like atropine, are not so readily absorbed and exert their effect only after injection (Wyman 1924).

The observed effects of the methonium compounds are somewhat confusing. The injection of decamethonium lightened fish which were dark. (See table.) This drug is reported to imitate acetylcholine very closely at the neuromuscular junction in some mammals (Paton and Zaimis 1952). On the other hand, the injection of hexamethonium darkened light fish, i.e., dispersed the melanophores. (See table.) This drug is said to have the capacity of "specifically antagonizing acetylcholine at the ganglionic synapse." Neither drug affected fish that were immersed. This might have been anticipated, because the methonium drugs are poorly absorbed by mouth or percutaneously. If much longer immersions had been tried, some effect might have been forthcoming. In any event, the absorption of methonium compounds by Fundulus during immersion is of low magnitude as compared with OMPA.

It is known that hexamethonium acts as a depressant on the central nervous system. Moreover, as a general rule drugs that depress the central or sympathetic nervous system in fish cause a dispersal of the melanophores and darken the fish (Wyman 1924). Fundulus possesses dineuronic melanophores. The pigment-concentrating fibers originate in specific loci within the central nervous system (Prosser 1950). Depression of these loci by drugs inhibits the action of the concentrating fibers and permits the darkening of the fish. The results of the hexamethonium injections in the present work are, therefore, explainable in terms of central nervous depression.

There are also present, in Fundulus, pigment-dispersing fibers that exert their action through the mediation of acetylcholine. Whereas hexamethonium exerts its action by blocking central ganglionic transmission, decamethonium is peripheral in its action. In mammals, it produces neuromuscular block through end-plate depolarization. The lightening of dark specimens of Fundulus after the injections of decamethonium is difficult to explain in terms of depolarization of the junction between the melanophore and the cholinergic pigment-dispersing fiber. Such a block should result in dark fish; the mechanism by which decamethonium lightens dark fish is still obscure.

It is interesting to note that both acetylcholine and atropine darken light fish. In mammals, atropine counteracts the effects of acetylcholine, within certain dosage ranges. In Fundulus, there is an apparent paradox which may have one of the following explanations:

1. No atropine antagonism toward acetylcholine at the melanophores.

2. Atropine block of the adrenergic ganglion.

The exact answer to this problem should be the object of future study.

Summary

1. Treatment of Fundulus with OMPA by injection or by immersion renders light specimens dark. The OMPA is probably transformed in the fish into an anticholinesterase product; the resulting accumulation of acetylcholine causes the darkening of the fish.

2. Injection of hexamethonium into a light Fundulus darkens the fish. This is explained on the basis of central nervous depression by the drug, resulting in the blocking of impulses via the adrenergic pigment-concentrating neurones.

3. Injection of decamethonium into a dark Fundulus renders the fish light. This result cannot be explained in terms of a peripheral block at the junction of the cholinergic pigment-dispersing neurone and the melanophore. The mechanism involved is still not clear.

4. The methonium compounds were not absorbed by fish immersed in them.

Acknowledgments

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The author wishes to express sincere gratitude to Dr. Herbert W. Graham, director of the Biological Laboratory, Bureau of Commercial Fisheries, Woods Hole, Massachusetts, for facilities and specimens that made this work possible.

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The American mission to the Russian salmon areas in the fall of 1959 had a twofold purpose: to give our specialists an opportunity to learn of Russian fishery operations at firsthand, and to secure fish and fish blood samples of known Russian origin for an international salmon study that has been in progress for about 3 years.

EXPERIMENTAL INTRODUCTIONS OF FRESH-WATER ALEWIVES

Robert E. Vincent
Department of Conservation, Cornell University
Ithaca, New York

DURING RECENT YEARS THE ALEWIFE (Alosa pseudoharengus Wilson) has received attention as a forage fish in fresh-water lakes. The annual transplanting of adult sea-run stocks into fresh-water ponds as forage for warm-water fish is receiving the most effort. Currently the practice is to stock mature fish and allow them to spawn, with the young providing immediate forage. All the alewives are utilized as food or die by the next spring. Two of Connecticut's several small lakes that contain landlocked alewives[1/] have not had free access to the ocean at some time in the past, the implication being that these lakes were planted with marine stock at some unknown time (James P. Galligan, personal communication). However, little attention has been paid to establishing these introductions as being self-sustaining.

Lake Ontario and certain of the Finger Lakes in central New York have supported populations of dwarf alewives since at least 1870 (Miller 1957). The Cayuga Lake dwarf stock has an average length of 5 inches, as compared with the 6 to 10 inches of the Lake Ontario alewives (Stone 1947) and 10 to 11 inches of the marine forms (Belding 1921). Although marine alewives are in current vogue as food for warm-water species, the dwarf fresh-water alewife has long been recognized as important forage for certain salmonids (Dymond 1928, Eaton 1928, Stone 1947, Royce 1951, Graham 1954). Problems associated with the experimental introduction of alewives into trout lakes of the Adirondack Mountains in northern New York are presented here. Cayuga Lake was the source of fish for these trials, which were conducted during the summer of 1958.

There were two objectives in undertaking this project: First, to find a practical means of transporting alewives as either eggs or adults; second, to determine how well alewives survive and reproduce in a foreign body of water.

Collection of Adults

Most fish were collected during evenings by seining gravel shoals with a nylon seine (75-foot, 0.25-inch mesh). A successful haul would catch 500 alewives. As an alternative method, a trap net was set for one night in the lagoonlike mouth of Taughannock Creek, one of the large tributaries of Cayuga Lake. The catch was nearly 1,000 alewives, but the immediate mortality due to fighting the webbing of the trap net was so great (23 percent) that this means of capture was considered impractical. Death of fish in the trap net, as well as throughout the handling process, was caused by cerebral hemorrhage in the frontal region. A peculiar skeletal structure makes the alewife susceptible to injury of this type. Thrashing of larger fish taken in the same seine as the alewives increased mortality by dislodging many of the alewives' extremely deciduous scales.

[1/] The fish in one lake might be Alosa aestivalis or possibly a hybrid with Alosa pseudoharengus.

Problems in Culturing

Artificially impregnating and incubating eggs, then transporting the eggs or fry appeared to be one possible method of introduction. Although Odell (1934) was successful in hatching alewife eggs in small numbers under laboratory conditions, the present investigation suggests that large-scale culture is not practical. Alewife ovaries maturate over a long period, never carrying a large percentage of ripe eggs at any one time. Under natural conditions this would induce intermittent spawning throughout the season. Even partly spawned females had green eggs still developing in the anterior portion of the ovary. This increases the difficulty of culturing large numbers of eggs, as many green eggs may be extruded with desired ripe ones. At the warm water temperatures (60° to 70° F.) needed for incubation, fungus develops rapidly. Belding (1921) noted that the principal difficulty with artificial hatching is acquiring mature females and ripe eggs.

Transportation

As artificial hatching seemed infeasible, transporting of adults was tried as an alternative. Attempts to haul marine alewives were made as early as 1921, when Belding found that 6 fish per 10-gallon can could be carried 8 miles without loss. Rounsefell and Stringer (1945), using a 500-gallon tank of 55° to 60° F. aerated water, had a 19-percent loss when carrying 300 to 700 fish per tank. Graham (1956) had 51 percent transport mortality when 576 adult Lake Ontario alewives were transported between June 16 and July 29.

After capture, the Cayuga alewives were transported 10 miles to the Cornell University Experimental Fish Hatchery, where they were put in standard hatchery troughs. The fish were held overnight before being hauled further because the peak of seining mortality is delayed 4 hours. Some fish held well in the 68° F. hatchery water for as long as 2 weeks.

To test the carrying capacity of 90-gallon, rectangular fish-distribution tanks, 200 (6 pounds), 300 (9 pounds), and 500 (5 pounds) fish were carried in each of the 3 tanks. Aeration was furnished by oxygen liberators at 10 pounds' pressure. Water temperature was between 65° and 70° F. After an 8-hour haul, mortality was approximately 10 percent in each tank. A 4-hour haul with 350 fish in each tank had a 6-percent loss.

Later in the season (after July 20), when the fish were partly spawned, hauling mortality increased sharply: 33 percent during a 4-hour trip and 90 percent during the following two trips. Reduction of the water temperature to 45° F. during transportation was ineffective in curtailing this late-season mortality.

A differential handling-and-hauling mortality was suggested by sexing 161 fish that died during transit. Of these, 89 were males and 72 were females, though there were 6.6 males to each female in 641 fish taken in Cayuga Lake in a 3-week period. The gravid females are more fragile than the males. Additional mortality due to handling could be expected after planting.

Results of Stocking

Four ponds of different biological and physical characteristics were planted with adult alewives early in the summer of 1958. The success of these plantings is reported here. A fifth planting--in Little Moose Lake, near Old Forge, New York--has not yet been evaluated.

Pond I. --This 0.2-acre, 5-foot-deep pond at the Cornell University Experimental Fish Hatchery contained no fish prior to stocking. A dense mat of Chara blanketed the pond bottom; plankton appeared to be sparse. On June 25, 100 male and 10 female alewives were liberated in the pond. A seine haul on August 19 recaptured 35 adults but no zero-age-class fish, indicating that survival of adults was good but reproduction was either lacking or slight.

Pond F. --This pond, near pond I and of similar size and depth, also contained no fish. Chara was not present in this pond. Fertilizer (100 pounds of 6-12-6) was added late in the spring, and the pond

had a good plankton bloom all summer. On June 25, 100 males and 42 females were stocked. A seine haul on August 19 took 6 adults and 60 zero-age-class fish. Adults were larger than when stocked; they averaged 5.9 inches (10.7 fish per pound), having grown from the prestocking mean length of 5.0 inches (33 fish per pound). The young were robust and averaged 1.7 inches in length. As the adults had been in the pond 55 days, the young could hardly be more than 40 days old.

In some areas, all fresh-water alewives die after spawning and the young fail to overwinter in small ponds. When pond F was drained on May 5, 1959, 70 planted adults and about 600 of their progeny were recovered. The young were 2.9 inches in mean length, having grown 1.2 inches since the previous August. Landlocked salmon 6 to 8 inches long were also in the pond over winter and were seen trying to feed on the young alewives. Immediately after the ice cover melted, the following recently dead fish were found: 3 adult alewives, 10 young alewives, and 4 salmon.

Chatfield Pond. --Located 4 miles east of Ithaca, New York, Chatfield Pond is cool, spring fed, and 0.2 acre in area. It supports a population of smallmouth bass (Micropterus dolomieu). Like pond I, this pond had a profuse growth of Chara. On June 27, 180 unsexed alewives were stocked; of these, 26 dead were removed in 2 days. A school of about 120 adult fish was seen July 4 and again July 12. When the pond was seined August 20, however, no adult or young alewives could be found.

Black Pond. --This 25-acre pond in the northern Adirondack Mountains is cool and deep (58 feet). It supported a varied fish population consisting of lake trout (Cristivomer namaycush), landlocked salmon (Salmo salar), kokanee (Oncorhynchus nerka), common sucker (Catostomus commersonni), common shiner (Notropis cornutus), bluntnose minnow (Pimephales notatus), northern redbelly dace (Chrosomus eos), and brown bullhead (Ictalurus nebulosus).

After an 8-hour trip, 900 alewives were planted in Black Pond on June 26. A trap net fished there in the last week of September to determine species composition caught 30 to 125 adult alewives in each 48-hour period. The pond was reclaimed with rotenone, October 3, 1958. Stocked adults and numerous progeny were recovered.

Fish from the original planting had a mean length of 6.3 inches (10 per pound), having grown 1.3 inches since stocking. The zero-age-class fish fell into two distinct length groups, one with a mean length of 3.0 inches (95 per pound) and a second with a mean length of 1.3 inches (1,500 per pound); sample sizes and ranges were 65 fish, 2.7 to 3.5 inches, and 67 fish, 1.1 to 1.7 inches, respectively. The bimodal size distribution indicated two successful spawnings. The larger size group seemed more abundant, perhaps only because the fish were more conspicuous; also, they appeared concentrated at the surface in an area of nearly an acre, suggesting the existence of at least one large school.

Discussion

With careful handling, fresh-water alewives can be transported to a different body of water and will spawn successfully. Seven fish per gallon of water were hauled several hundred miles; as this was the maximum density attempted, it is possible that more fish per gallon could be transported without undue loss. A sudden increase in mortality as spawning begins limits the time during which fish can be transported. It seems necessary, therefore, to transport alewives early in the season when they first appear on the spawning shoals. Greater susceptibility to handling mortality can be expected as the season advances, for midsummer die-offs are common. The inability of alewives to acclimate to the warm water of the shallows was given by Graham (1956) as the reason for summer mortality. Hoar (1952) suggested that an exhausted thyroid is the triggering mechanism for this annual weakness and mortality. As all the fish used in these tests were taken when the lake surface temperature was 66° or 67° F., it does not seem that high temperatures caused the late-season mortality associated with transportation.

There was no natural spawning in ponds that contained excessive growths of Chara. This ecological condition may have inhibited spawning or survival of eggs and young. Although prolonged spawning is a hindrance to artificial incubation, it is a desirable characteristic for a forage fish.

Satisfactory growth and reproduction in two such widely different ponds indicate the versatility of alewives.

Acknowledgments

This project was undertaken as part of long-term fishery-management studies by Cornell University in the Adirondack Mountains of New York.

The advice of Dr. Dwight A. Webster, professor of fishery biology at Cornell University, and his review of the manuscript are much appreciated. The assistance of Mr. B. E. Brown and others who helped with seining operations, and the cooperation of the Adirondack League Club of Old Forge, N. Y., and Mr. and Mrs. D. P. Ross of Montchanin, Del., in permitting the use of ponds and facilities, are gratefully acknowledged.

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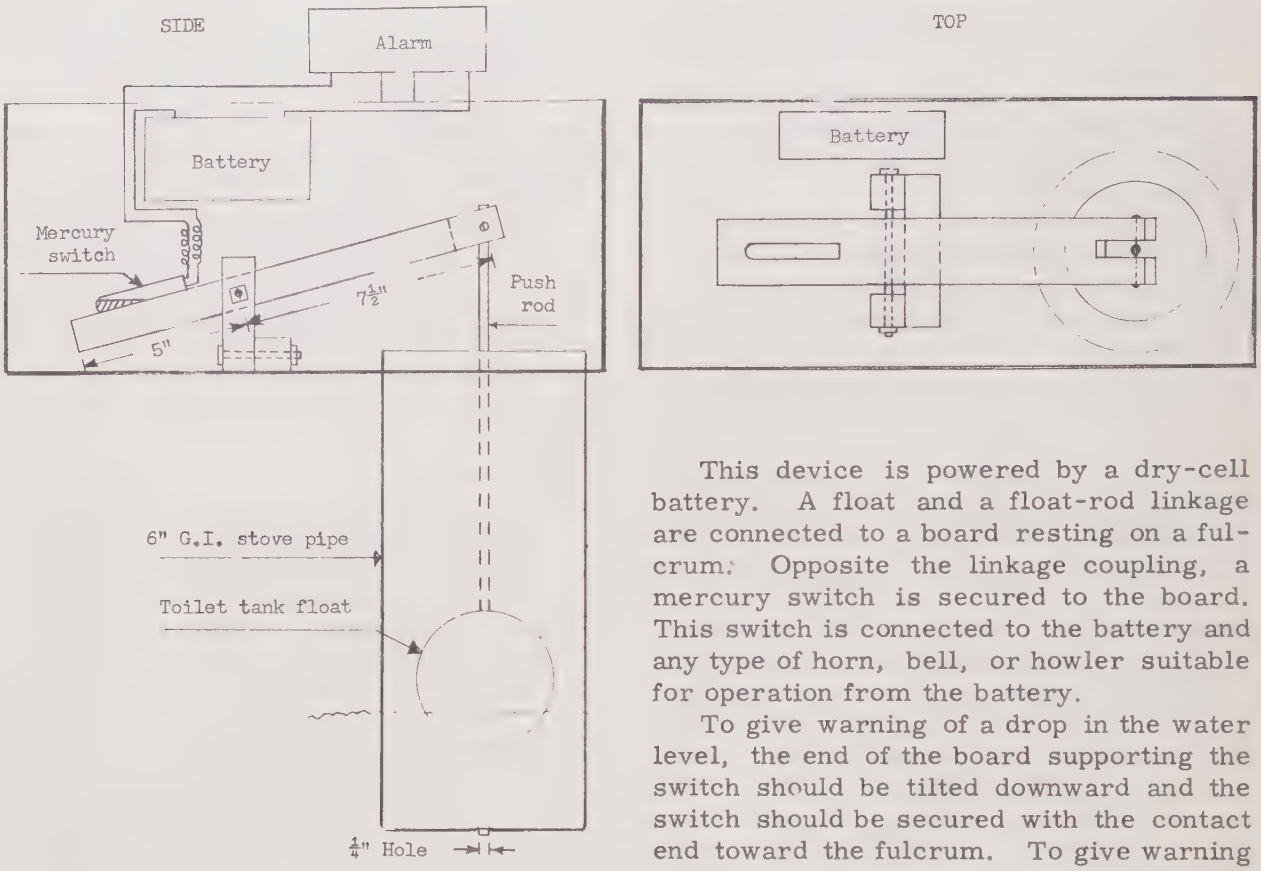
WARNING DEVICE FOR USE AT FISH HATCHERIES



At any hatchery, an unexpected or unobserved rise or fall in the water level may result in a considerable loss of fish. To provide a warning of any such changes in water levels, a portable self-contained alarm apparatus has recently been devised at the National Fish Hatchery at Carson, Washington.

This device is powered by a dry-cell battery. A float and a float-rod linkage are connected to a board resting on a fulcrum. Opposite the linkage coupling, a mercury switch is secured to the board. This switch is connected to the battery and any type of horn, bell, or howler suitable for operation from the battery.

To give warning of a drop in the water level, the end of the board supporting the switch should be tilted downward and the switch should be secured with the contact end toward the fulcrum. To give warning of a rise in the water level, the switch should be secured in the reverse position and that end of the board should be tilted upward.

The permissible variation in water level determines the length of the board.

---VICTOR T. REED, Bureau of Sport Fisheries and Wildlife, Carson, Washington.

Myxosoma (Lentospora) cerebralis apparently causes "whirling disease" in salmonids.

OUTBREAKS OF COLUMNARIS IN CENTER HILL AND OLD HICKORY RESERVOIRS, TENNESSEE

Billy G. Isom
Stream Pollution Control, Tennessee Department of Public Health
Nashville, Tennessee

AN EPIDEMIC THAT RESULTED IN GREAT MORTALITY AMONG THE GAME FISHES occurred on Center Hill Reservoir in DeKalb, White, Warren, and Putnam Counties, Tennessee. The reservoir dam is at mile 26.6 on the Caney Fork River, 21 miles from Cookeville and 17 miles from Carthage, Tennessee. The reservoir has a surface area of 18,220 acres at elevation 648. Its primary purposes are flood control and power production. The reservoir is in a predominately agricultural area, but the lake shores are dominated by wooded hills. Since its completion the reservoir has been a valuable recreational area, especially for sport fishing.

About a week after the fish kill in Center Hill Reservoir, there was another fish kill in the downstream Old Hickory Reservoir.

Field Investigation

On May 7, 1959, an epidemic fish disease in Center Hill Reservoir was investigated. Reports of fish mortality throughout the reservoir had been common 3 weeks prior to this investigation. Inquiry revealed that this reservoir received no domestic or industrial pollution of any consequence.

The investigation revealed many dead and moribund fish. Gross examination of the fish revealed rare (if any) hyperplasia, white spots, necrotic areas, frayed fins, and usually a secondary invasion of the tissues by a fungus.

The species affected were principally the black basses (Micropterus salmoides and M. dolomieu), the bluegill (Lepomis macrochirus), and white crappie (Pomoxis annularis). It was estimated that 80 to 90 percent of the dead fish were black bass.

The findings with regard to susceptibility of species are contrary to any yet reported. Davis, in reference to Cytophaga columnaris, wrote (1953: 266):

> ... Among the more susceptible species are the buffalofishes (Ictiobus bubalus and Megastomatobus cyprinella) and the crappies (Pomoxis annularis and P. nigro-maculatus). The less susceptible species include the black basses (Micropterus salmoides and M. dolomieu), the channel catfish (Ictalurus lacustris), and various species of sunfishes. ...

Scaleless warm-water fish are considered particularly susceptible (Snieszko 1958). Of the species considered more susceptible to this pathogen, only P. annularis was noted.

The records of the U. S. Corps of Engineers show that on April 30, 1959, the temperature of the water at the surface was 64° F., and that on the following day it had risen to 73° F., an increase of 9° in 24 hours. (See figure.) According to reports, more fish were noted to be dead or dying on May 2-3, 1959, than at any other time. By May 7 the epidemic had abated, and few mortalities have been reported since.

The fish kill caused by this pathogen on Old Hickory Reservoir was investigated

Surface water temperature, Center Hill Reservoir, 1959.

on May 15, 1959. The investigation revealed that white crappies (P. annularis) constituted about 85 to 90 percent of the fish affected. Other species affected in this reservoir were M. salmoides, Dorosoma cepedianum, Ictalurus furcatus, and Cyprinus carpio. In this study the pathogen was isolated from a white crappie.

Laboratory

Instructions provided by the Microbiological Laboratory, Bureau of Sport Fisheries and Wildlife, at Kearneysville, West Virginia, were used in isolating the bacterium. Ordal and Rucker reported (1944: 16) that "a medium containing 0.25% to 0.50% Bacto-tryptone and 0.5% to 0.9% agar adjusted to pH 7.3 was found to be very satisfactory for isolation and cultivation." This medium was employed to isolate Chondrococcus columnaris.[1/] Our findings were confirmed by Lyle S. Pettijohn, fishery research biologist, Eastern Fish Disease Laboratory, Bureau of Sport Fisheries and Wildlife, at Kearneysville, West Virginia. Fish and Rucker (1945) isolated the pathogen from internal organs of salmonid fishes. No references were found on the isolation of this pathogen from either L. macrochirus, M. salmoides, or M. dolomieu inhabiting natural waters. Isolation was made from the kidney of a bluegill taken in a moribund state.

Experiments showed that the isolated pathogen was lethal to L. macrochirus at 26° C. The optimum temperature range was not determined. Garnjobst (1945) found that the optimum temperature range for Cytophaga columnaris was from 25° to 30° C. Ordal and Rucker wrote (1944: 16):

> Transmission experiments under controlled conditions have shown the myxobacteria to be pathogenic to salmonid fishes. The mortality, however, depended upon the temperature at which the fish were held. ...

Experiments were conducted with the water in which the experimentally affected fish were held. The pathogen was isolated from the water by loop transfers and with a membrane filter. Isolation was made from 0.1 milliliter of the water with the membrane filter. This was the smallest quantity of water used. In other experiments with this water, the pathogen was not transmitted to healthy L. macrochirus.

In transmission experiments with the cultured pathogen, transmission was via the oral cavity, a dorsal slit, and gill filaments. Seventy-five percent of the test specimens died in all three administrations.

Acknowledgments

Sincere appreciation is expressed to C. E. Ruhr and other personnel of the Tennessee Game and Fish Commission; Harold N. Mullican and Ralph M. Sinclair, Tennessee Stream Pollution Control Board; Martin D. Wolfe, bacteriologist, Tennessee Department of Public Health; and especially Dr. S. F. Snieszko and Lyle Pettijohn, Eastern Fish Disease Laboratory, Bureau of Sport Fisheries and Wildlife, Kearneysville, West Virginia, for their most kind assistance, suggestions, and criticisms.

1/ As production of microcysts in culture media was not established, it is possible that the myxobacterium isolated should be classified as Cytophaga columnaris. This point is difficult to resolve because production of microcysts is not a stable characteristic (Rucker, Earp, and Ordal, 1954).

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FISH MORTALITIES ASSOCIATED WITH CLOUDY WEATHER

Heavy fish mortalities that occurred in a single night in five small ponds (0.5 to 1.5 surface acres) in Davidson County, Tennessee, were reported on August 14, 1954. These mortalities were probably only a small sample of those that actually occurred that night: the presence of dead fish in other ponds was reported in the following week, but the exact date of those mortalities was not known. All ponds had been stocked with bluegills and largemouth bass; several also had black bullheads and green sunfish. Most of the dead fish were large bluegills and largemouth bass. None of the ponds suffered complete mortalities.

Data on air temperatures and sunlight were taken from U. S. Weather Bureau records at Berry Field, within 5 miles of all the ponds. These records showed that a cloudy period preceded by several days of high temperature was associated, at least, with these mortalities.

Other observations in ponds near Nashville showed that when water temperatures exceeded 95° F., oxygen concentrations of 16 to 20 p.p.m. (over 200 percent saturation) could often be measured between 3 and 4 p.m. Presumably, oxygen readings would have been low the day before these mortalities occurred, indicating an upset in the balance between photosynthetic activity and algal decomposition and respiration. By watching weather reports and taking oxygen readings in midafternoon in cloudy weather, warm-water hatchery managers might be able to foresee--and prevent--such mortalities.

MAXIMUM AND MINIMUM AIR TEMPERATURES AND MINUTES OF SUNLIGHT, August 10 - 16, 1954, AT BERRY FIELD, DAVIDSON COUNTY, TENNESSEE

August	Temperatures, °F. Maximum	Minimum	Minutes of sunlight
10	93	62	810
11	89	67	703
12	72	67	0
13	87	66	107
	Fish mortalities		
14	99	69	745
15	101	77	792
16	103	74	699

--- NORMAN G. BENSON, Bureau of Sport Fisheries and Wildlife, Logan, Utah.

COMMENTS

The exchange of comments appearing here was inspired by the publication of A. N. Woodall's paper, "The Missing Links of Fish Husbandry" (THE PROGRESSIVE FISH-CULTURIST 21 (2): 51-54). Introductory and closing remarks not important to the argument have been deleted.--The EDITOR.

Bureau of Sport Fisheries and Wildlife
Eastern Fish Disease Laboratory
Kearneysville, West Virginia

July 15, 1959

I have read with great interest the timely and lucid article, "The Missing Links of Fish Husbandry" by Mr. Woodall. I agree with him that in research on fish diseases and nutrition there is a need for "pilot-plant studies wherein the test tube results of basic research are adapted to production problems."

Fish hatcheries designed and operated exclusively as pilot plants would be desirable. In a large measure, however, the existing fish hatcheries can serve for this purpose at least until special pilot-plant hatcheries are built. It must be added here that some existing Federal and State trout hatcheries have been constructed to serve for production, as "pilot plants," and for research. Much important information on feeding of trout, raising of warm-water fish, and on practical value of disease-control measures has been obtained at such hatcheries.

As I see it, the Federal fish hatcheries need not be used exclusively for raising fish. They can be useful as model hatcheries and pilot plants. This should in no way reduce their value for fish production, because to be model hatcheries they must grow fish efficiently. The negative attitude toward research by "practical" fish-culturists is disappearing. I have learned through contacts with personnel in many Federal and State hatcheries, with management biologists, and with supervisors of fish culture, that there is a receptive attitude toward use of fish hatcheries for experiments which can be run with existing facilities and personnel without any serious impairment to fish production.

I am not so familiar with current use of State hatcheries as pilot plants, but I know that at some Federal hatcheries pilot-plant experiments are being made on control of diseases and use of commercial trout feeds. For example, at the Berlin, New Hampshire, trout hatchery experiments were conducted on the treatment of kidney disease [1/] and, for the past 2 years, have been conducted on the effect of the control of external parasites on the incidence of kidney disease. In this second experiment thousands of fish have been examined by the station personnel and prepared for staining and examination at the Eastern Fish Disease Laboratory, and weekly formalin treatments have been given. This has required a prodigious amount of work by hatchery personnel. Similar experiments on the control of kidney disease have been conducted at trout hatcheries in the southeastern States.

Another arrangement in trout production hatcheries is for the evaluation of pelleted fish diets. This type of work can be done well at all modern trout

1/ Snieszko, S. F., and P. J. Griffin. 1955. Kidney disease in brook trout and its treatment. Prog. Fish-Cult. 17 (1): 3-13.

hatcheries. Most hatcheries have facilities for refrigeration of fresh meat and storage space for pellets.

I know of some pilot-plant experiments, or field tests, that are being conducted in hatcheries in cooperation with research laboratories. Some other experiments have not been adequately planned or conducted, but means to remedy this situation are readily available.

If pilot-plant experiments are well planned, only a small part of the production facilities of a hatchery need be used for testing. It is mandatory, however, that tests be made in replicate, that the test fish be uniform in every respect, and that all conditions in the treated and control groups be the same except for the treatment or the test diet.

It is of the utmost importance that only well-planned experiments be conducted, that conditions at the selected hatcheries permit simultaneous replication of tests, and that adequate controls be included. One should keep in mind that conclusions derived from incorrectly planned and executed experiments may do more harm than no experiments at all.

-- S. F. Snieszko

Bureau of Sport Fisheries and Wildlife
Western Fish Nutrition Laboratory
Cook, Washington

July 24, 1959

What Dr. Snieszko is discussing is the final step of a pilot-plant operation--the testing of a scientifically developed diet on a production scale. I repeat, this is the final step, practically the fruit of the pilot plant's labors. Before this step can be taken, the real work of the pilot plant must be done. This, of course, includes chemical, microbiological, and biological assays of dietary ingredients, as well as the determination of digestibility coefficients and biological values and the effect of varied environmental conditions (water temperature and chemistry) on the same. After these analytical procedures will come the compounding and testing of a multitude of diets under closely controlled and varied conditions on a true pilot-plant scale. This is applied and developmental research, and from it will come the knowledge necessary to the development of diets suitable for specific hatchery conditions. To be sure, the diets will then have to be carefully tested and evaluated under production conditions so that, if necessary, further modifications can be made.

This all reemphasizes my major point: our knowledge at present is insufficient to conduct this type of experiment with any assurance of success or even adequate means of evaluation. This knowledge can and must be supplied by the preliminary pilot-plant research previously mentioned.

Dr. Snieszko refers to adequate pilot-plant studies which were conducted at the Berlin hatchery. I should be very much surprised indeed if these studies were made prior to his own basic research on the disease and prior to his applied and developmental (pilot-plant) research, which probably included tests on a trough basis in his hatchery.

This question will surely arise: If these pilot-plant studies are research, why then don't we do them? Obviously, the answer to this is that we can. However, with limited personnel and facilities, each applied problem we undertake will limit the amount of basic research we can do. The amount of applied or developmental pilot-plant research now needed is such that if we were to do it, we would have no time for basic research. As indicated in my paper, the interdependence of the problem is such that one cannot even adequately design or conduct the preliminary pilot-plant studies until that basic research on which the particular pilot-plant study depends is done. Basic research must, therefore, keep well ahead of and anticipate the needs of the pilot plant. Beyond all question, the initial pilot-plant studies must be completed before diets can be compounded. Furthermore, reason and prudence dictate that initial testing should be done with no more fish than one can afford to sacrifice.

To summarize: Is the link still missing? -- A resounding yes! Can this missing link be supplied by hatchery testing under the direction and supervision of a committee (or anyone else, for that matter)? -- Most emphatically, no.

The fact that these questions have been raised by a scientist of Dr. Snieszko's stature points out how painfully inadequate my article was in achieving its objective--that of describing the interrelationship of basic research, pilot plant, and quality control. Also, I wanted to leave a clear understanding as to the functions of the pilot plant and quality-control laboratory, the complexities of the problems undertaken, and, therefore, the type of people needed for its operation. Above all else, I wanted to show how sound nutrition practices can and will be achieved through a rational, methodical, stepwise approach. These things, although briefly stated, seemed clear to me and to my associates here who read the article.

So, what is to be done? Although I admit the failure of "The Missing Links" to achieve its objectives, I feel that the attainment of these links is so important that I must refuse to admit to perpetual defeat.

-- A. N. Woodall

Bureau of Sport Fisheries and Wildlife
Eastern Fish Disease Laboratory
Kearneysville, West Virginia

August 3, 1959

I must admit that I was surprised by Mr. Woodall's reaction to my comments, as I had nothing but praise for his paper, "The Missing Links of Fish Husbandry." My complete agreement with his thesis was expressed in the first sentence of my second paragraph: "Fish hatcheries designed and operated exclusively as pilot plants would be desirable." This statement I then qualified as follows: "In a large measure, however, the existing fish hatcheries can serve for this purpose at least until special pilot-plant hatcheries are built."

I am convinced that it is not necessary to wait for the construction of special pilot plants, because many types of pilot-plant experiments can be conducted with facilities already existing at some--but not all--fish hatcheries. I agree with Mr. Woodall's statement (July 24, 1959) that I had in mind only "the final step of a pilot-plant operation." I believe that fundamental and developmental research should be conducted at research laboratories or research hatcheries--or whatever one may wish to name them--but not in pilot plants. Quality control is another subject, which I do not wish to discuss here.

I have not even implied, in my comments regarding the paper, that my ideas should replace Mr. Woodall's; I have merely made some practical suggestions based on my experience.

Fish-husbandry problems are so many and so varied that forging the missing links must be the business of all concerned. Pilot plants and quality control may not seem so remote and improbable since the publication of this article and this exchange of views.

-- S. F. Snieszko

ALGAE IN WATER SUPPLIES, AN ILLUSTRATED MANUAL ON THE IDENTIFICATION, SIGNIFICANCE, AND CONTROL OF ALGAE IN WATER SUPPLIES, by C. Mervin Palmer of the Robert A. Taft Sanitary Engineering Center, Public Health Service, U. S. Department of Health, Education and Welfare, is for sale as Public Health Service Publication No. 657 by the Superintendent of Documents, Government Printing Office, Washington 25, D. C., for $1 a copy.

TO CONTRIBUTORS

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